CEREAL CHEMISTRY

VOL. XV JULY, 1938 No. 4

SOME RESULTS WITH A NEW RECORDING MIXER FOR USE WITH SMALL SAMPLES [1]

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(Read at the Annual Meeting, May 1937)

Introduction

There is urgent need for a reliable test for baking quality that can be used by plant breeders in the early stages of the development of their new strains of wheat. The quantity of grain available precludes the use of milling and baking tests or of dough-testing machines such as the Brabender, Swanson, Chopin or Buhler. The dough-ball test uses only small quantities of material but it is far from precise.

Considering the three general factors controlling baking quality, protein content and gassing power can be readily determined on small samples but gluten quality can only be inferred from gluten washing, baking tests or mechanical tests. The determination of physical properties seemed to offer the greatest promise of adaptability to small samples. Accordingly a recording mixer, operating with samples containing 7 g. of dry matter, was designed and constructed. Preliminary tests of its accuracy and utility are reported.

Description of Mixer

A general view of the machine is shown in Figure 1. There is a stainless steel spindle mounted on a vertical shaft, driven at 120 R.P.M. by a 1/20 H.P. synchronous motor. This spindle rotates inside a water-jacketed stainless steel cup which is free to rotate against spring tension. Figures 2 and 3 show the cup and spindle in greater detail. The spindle has three curved outer pins and a straight centre pin while the cup has two curved pins which are located between the centre pin and the outer pins of the spindle when the mixer is assembled. The purpose of the curvature of the pins is two-fold:

[1] Published as Paper No. 132 of the Associate Committee on Grain Research of the National Research Council of Canada and the Dominion Department of Agriculture.

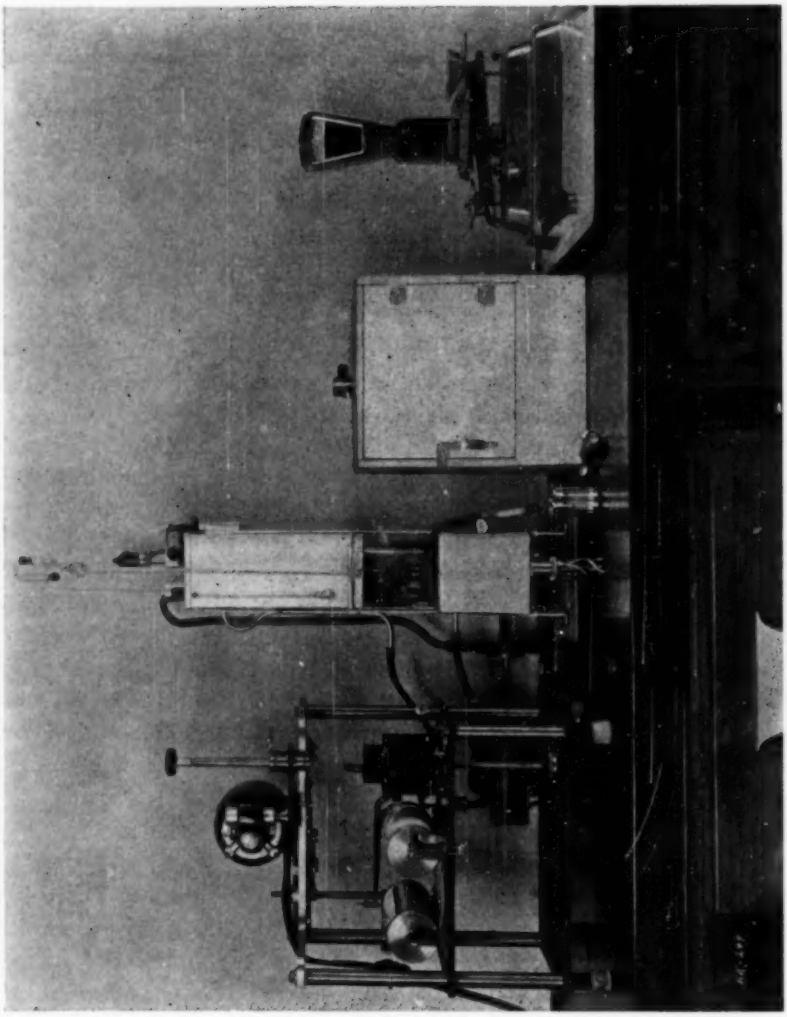

Figure 1. Recording mixer and auxiliary equipment.

(1) to keep the dough down to the bottom of the cup, and (2) to reduce the jerky action that intermittent passing of straight pins would produce.

In the model now in use the spindle was cut by hand from a stainless steel cylinder and the cup-pins were bent from stainless steel rod with the aid of a jig. The spindle can be reproduced accurately by

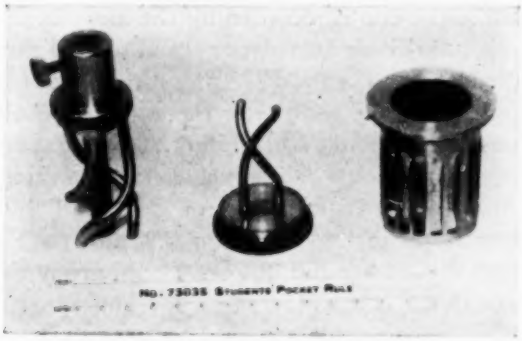

Figure 2. Spindle and cup.

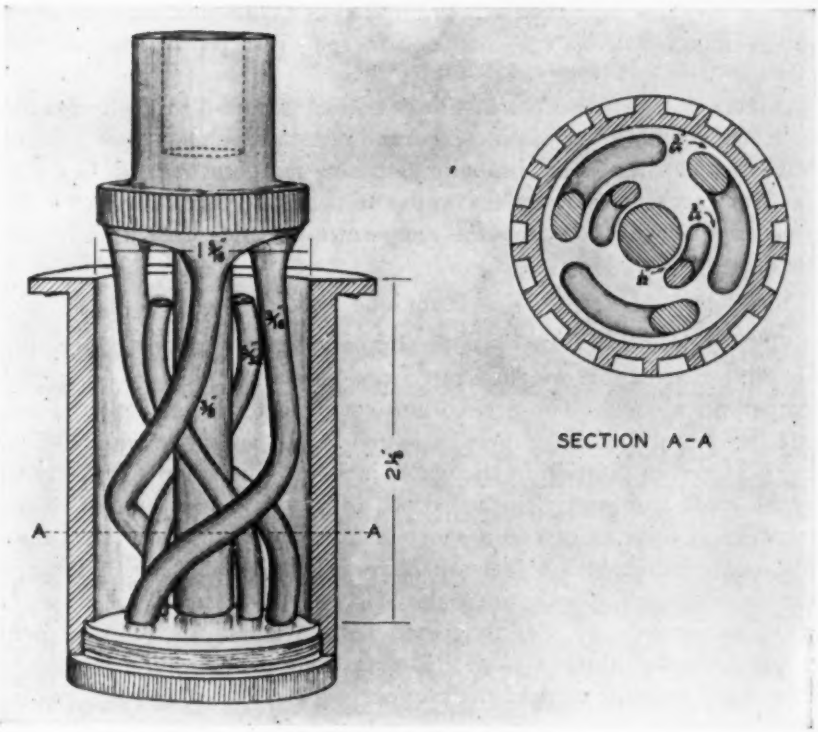

Figure 3. Spindle and cup assembly.

hand but difficulty has been experienced in bending the cup-pins to constant curvature. It is intended to modify slightly the shape of both the cup and the spindle pins to permit machine manufacture. The cup was machined from a solid piece of stainless steel and is provided with external ribs to improve the heat transfer to the jacketing water. To facilitate cleaning, it has a removable bottom machined to a water-tight fit.

The rotation of the cup is recorded by the movement of a pen arm on a kymograph roll driven at a paper speed of 0.44 mm. per second by a ratchet device from the motor. The torque, calculated as being exerted in the plane of the centres of the curved spindle pins, has a minimum value of 1225 g. and a maximum of 7500 g. with the present setting of the spring tension. These values may be raised or lowered, or the range between them increased or decreased, by altering the spring tension or by changing the springs, within the limits imposed by the power of the motor and the structural strength of the pins. With the present range, a pen movement of 1 mm. represents a change of 44.65 g. Plain paper strips are used for recording and the values read off by means of a scale scratched on celluloid. In the preliminary experiments the scale used was graduated in millimeters on both axes, but a change will be made to one reading directly in seconds and grams.

Figure 1 shows also the auxiliary equipment used in the tests: the scale for weighing the samples, a small constant temperature cabinet in which the samples are brought to mixing temperature (30° C.), and the apparatus which supplies water at constant temperature to the cup jacket and also controls the temperature of the water used to mix the doughs.

Technique

The technique of the test is simple. Flour samples containing 7 g. dry matter are weighed out from larger samples stored in the tempering cabinet. The desired amount of water is run into the cup from the burette and the flour is poured in on top of the water. The cup is placed in position in the water jacket. The spindle is attached to the shaft and gently pushed down into position in the cup. The kymograph drive is turned a short distance by hand to mark a base line for measurements and the machine is started. At the end of the run the cup and spindle are removed, washed under a jet of water at approximately 30° C., and dried with a clean towel. One man regularly makes about 8 single tests an hour including all operations except the measurement of the curves.

Character of Curves

The curves obtained differ in several respects and some of the types are shown in Figure 4. The height and time of occurrence of the maximum vary, as does the shape of the curve from the start of mixing to the maximum. There is a break in the curvature of the line some

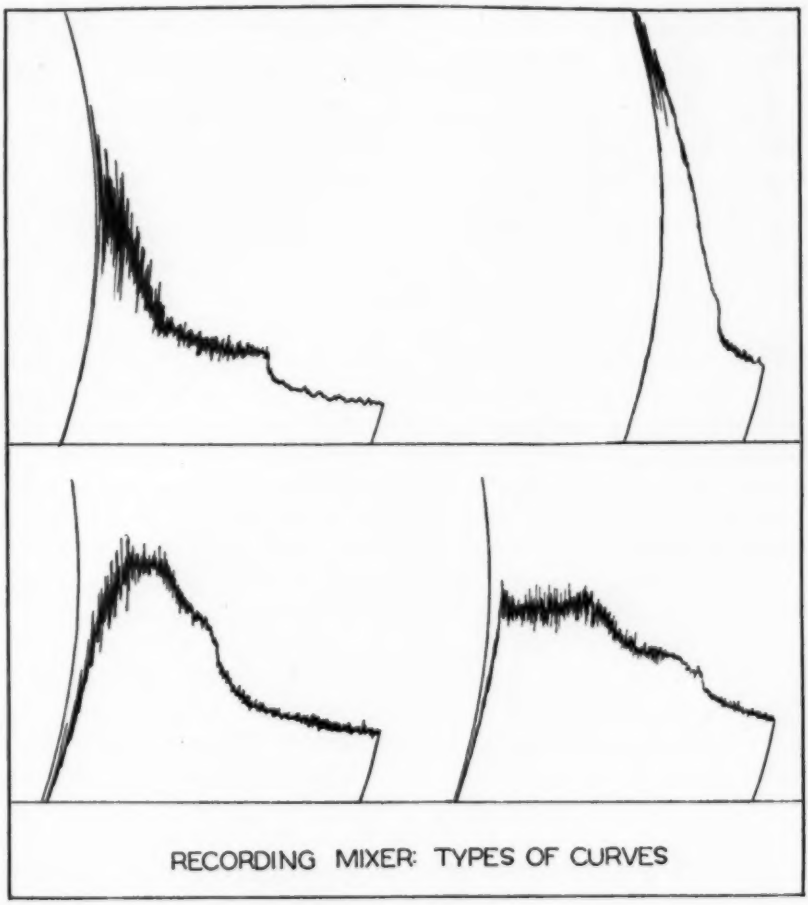

Figure 4. Types of curves.

time after the maximum, the position and sharpness of which are characteristic of different flours and treatments. The slopes and widths of line both before and after the break also vary. Each of these characters will have to be investigated in detail before the significance of the curves in terms of dough characteristics can be completely determined.

The curve can be quite accurately described by the height and time of the maximum, the height and time of the break, the angle at this point and by equations for the portions of the curve before and after the break. Equations for typical curves are shown in Table I.

TABLE I

EQUATIONS FOR PORTIONS OF CURVES BEFORE AND AFTER THE BREAK

Flour	Before break [1]	After break [2]
A	$H = 1.46\ T^{1.45}$	$T = .51\ e^{.12\ H}$
B	$H = 1.64\ T^{1.30}$	$H = .53\ T^{1.46}$
C	$H = .8 + 1.1\ T$	$T = 2.34\ e^{.03\ H}$
D	$H = -2.0 + 2\ T$	$H = .57\ T^{1.48}$
E	$H = .17\ T^{1.3}$	$T = 1.31\ e^{.22\ H}$
F	$H = 2.16\ e^{.237\ T}$	$T = .658\ e^{.139\ H}$
G	$H = 1.62\ e^{.11\ T}$	$H = .9 + 2.67\ T - 0.84\ T^2$
H	$H = 2.27\ T^{1.40}$	$H = 1.82\ T^{.43}$

[1] H = distance above the break; T = time before the break.
[2] H = distance below the break; T = time after the break.

Preliminary experiments have shown that the height of the maximum is affected by the total water in the dough (water in flour + water added) but that the moisture content of the flour and hence the amount of water added to bring the dough to constant water content also have an effect. It is probably affected also by the grade of the flour.

So far it has been possible to make a detailed study of only one feature of the curves, namely, the break. The position of the break is reproducible in replicate samples with a surprising degree of accuracy. Typical examples of the checks between triplicate determinations of the height and time of occurrence (measured in millimeters) are given in Table II, but a more reliable estimate of the reproducibility of these measurements can be obtained from the standard errors derived from 48 sets of triplicate determinations. These standard errors are 1.46 mm. for the height of the breaking point and 0.88 mm. for

TABLE II

REPLICABILITY OF POSITION OF BREAKING POINT

	Height, mm.			Time, mm.		
Flour	1	2	3	1	2	3
A	55	58	57	27	27	27
B	48	49	49	30	30	28
C	33	33	33	30	30	30
D	19	21	20	20	21	21
E	13	14	16	43	46	45
Standard Error (144 curves)		1.46 mm.			0.88 mm.	

the time of breaking. In other words, differences between the means of duplicate determinations are statistically significant if they exceed 3 mm. for height or 2 mm. for time of breakdown. As the height may vary from 10 mm. to 120 mm. and the time over about the same range, it is hardly possible that such a level of reproducibility could be obtained unless the measurements had some definite relation to dough character.

Significance of Height of Break

Figure 5 shows the enormous difference in height when two samples of flour with different protein contents are doughed with equal amounts of water, while Figure 6 shows the effect of varying the amount of

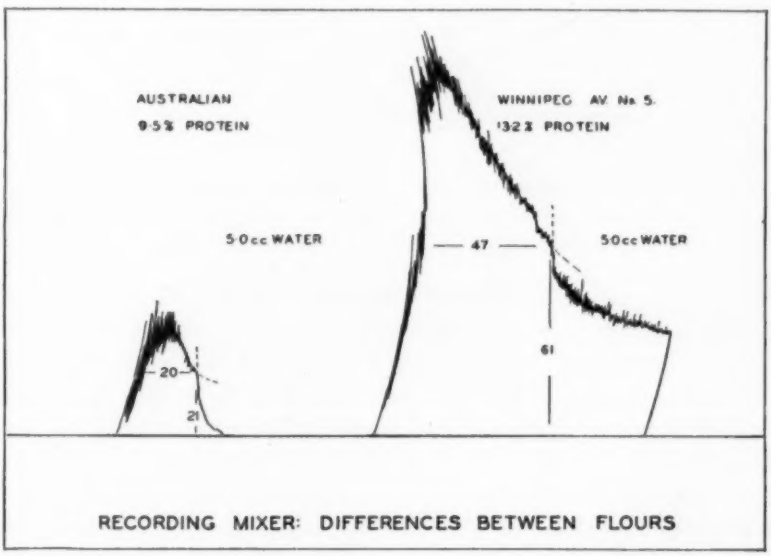

Figure 5. Curves from flours of different protein content.

water added. Other factors that probably influence the height of the break are: the grade of the flour, the granulation, the moisture content of the sample and the soundness of the wheat from which the flour was milled. So far, however, no direct experiments have been made on the effect of these factors.

The variation with protein content is important because it links one of the characteristics of the curves with a constituent of the flour. In a series of four varieties of wheat grown at seven different points in Alberta the simple correlation between protein content and the height of the break was $r = 0.71$ (1% point = 0.49) when the samples

were doughed with a constant amount of water, and an earlier series gave an even higher coefficient ($r = 0.88$, 1% point = 0.49).

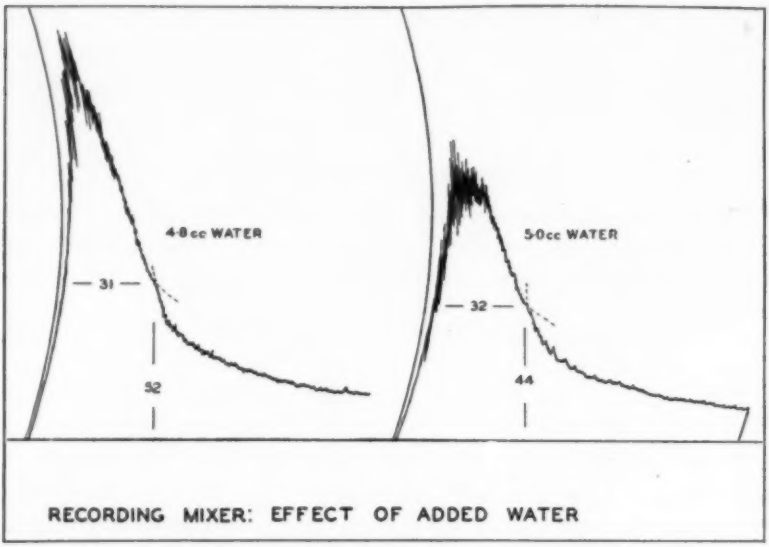

Figure 6. Effect of variation in amount of water added.

Significance of Sharpness of Break

In the Alberta series, mentioned above, it was noticed that there was considerable variation in the sharpness of the break. The angle between the slope of the lines before and after the break was measured for each curve so that increasing angle indicated increasing sharpness of break. This angle was correlated with protein content and the coefficient was found to be $r = 0.90$ (1% point = 0.49). As with the correlation reported in the last section, the importance of this lies in the establishment of a definite relation between the mixer curves and one of the factors of baking quality, rather than in the possibility of estimating protein content from the curves. The Kjeldahl determination is a simpler method of obtaining this information.

The sharpness of the break is influenced also by the amount of water added but except at the extremes of the range of the machine the change is comparatively small. In the Alberta series the average change in angle for an increase of 3% in absorption was only slightly over 2° while the range between flours was 50°.

Significance of Time of Occurrence of Break

Variations in the breaking time from about 35 seconds (16 mm.) to about 250 seconds (118 mm.) have been obtained with different

flours. The difference between two flours can be readily seen in Figure 5.

Where different amounts of water are added to the same flour (Figure 6) the breaking time is practically unaffected. This is shown for a number of flours in Table III. Curves were also obtained from 16 flours, to each of which three different amounts of water were added and the determinations made in triplicate giving nine results for each flour. Standard deviations were calculated: (1) regarding the nine curves from each as replicates in spite of the variations in water added and (2) in the ordinary manner from the triplicates. The former was 1.44 mm. and the latter 0.88 mm., indicating that the variation in water added made little difference to the breaking time. Later work has confirmed this conclusion.

TABLE III

EFFECT OF AMOUNT OF WATER ADDED ON BREAKING TIME

Water added	Breaking time, mm.				
	A	B	C	D	E
Control	42.3	26.7	40.0	58.7	18.7
+ 0.2 cc.	43.0	27.3	40.3	57.3	20.0
+ 0.4 cc.	43.3	29.7	40.3	56.0	20.3

Standard Error (144 curves)—at a single moisture 0.88 mm.
—neglecting moisture 1.44 mm.

This result was so striking and unexpected that further experiments were performed to test the constancy of the breaking time under different conditions. In the first experiment samples were diluted with 25% of starch without affecting the breaking time. Figure 7 is typical of the results obtained. Curves were also made with samples composed of starch mixed in various proportions with dried gluten.[2] The results, given in Table IV, again show the breaking time to be constant.

TABLE IV

EFFECT ON BREAKING TIME OF PROPORTION OF STARCH USED TO DILUTE DRIED GLUTEN

Dilution		Breaking time, mm.						
Gluten, %	Starch, %	A	B	C	D	E	F	G
20	80	56	24	18	38	31	32	24
15	85	56	24	18	40	30	31	24
10	90	56	26	17	40	32	36	22

[2] These gluten samples were prepared by T. R. Aitken of the Laboratory of the Board of Grain Commissioners, Winnipeg.

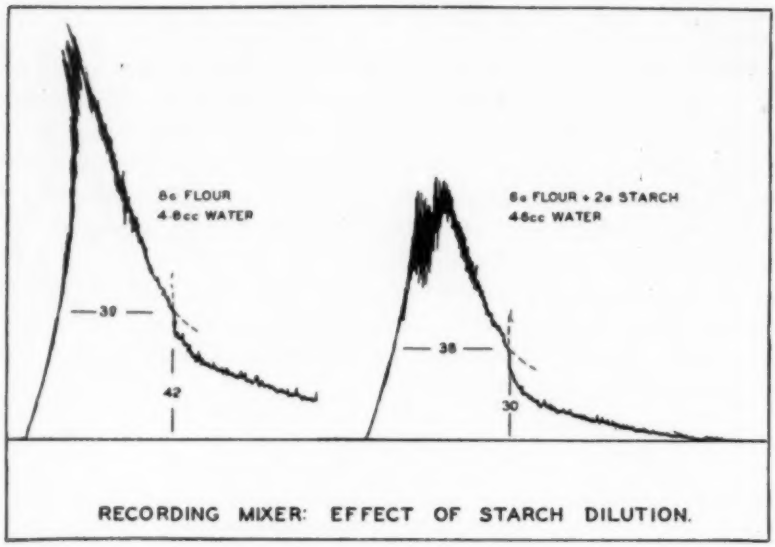

Figure 7. Effect of addition of starch.

Still another experiment was performed in which the weight of sample was varied from 7 g. to 9 g. (as received, moisture basis) and again the breaking time was unaffected (Table V).

TABLE V

EFFECT OF WEIGHT OF SAMPLE ON BREAKING TIME

Weight, g.	Breaking time, mm.		
	Flour A	Flour B	Flour C
7.0	34.5	42.0	37.0
7.5	34.5	42.0	36.0
8.0	36.5	41.5	38.0
8.5	36.0	42.5	39.5
9.0	—	43.5	38.5

It is apparent that the breaking time is characteristic of the flour. It is independent of the protein content and of the amount of water added except when excessive amounts of water are added or the protein content is abnormally low. If the protein content is reduced below a point not yet accurately determined but in the neighborhood of 7%, the breaking time decreases with increase in the water added.

To obtain preliminary information on the meaning of the breaking time in terms of flour characteristics, reagents known to affect colloidal properties were added to the doughs. Sodium chloride, lactic acid and ethyl alcohol were used and the results with one flour are shown in Figure 8. All of these reagents have a marked effect on the breaking

time, which therefore seems to be a measure of some colloidal property or properties. It is probably not a measure of the hydration capacity since lactic acid, which should increase hydration, and alcohol, which

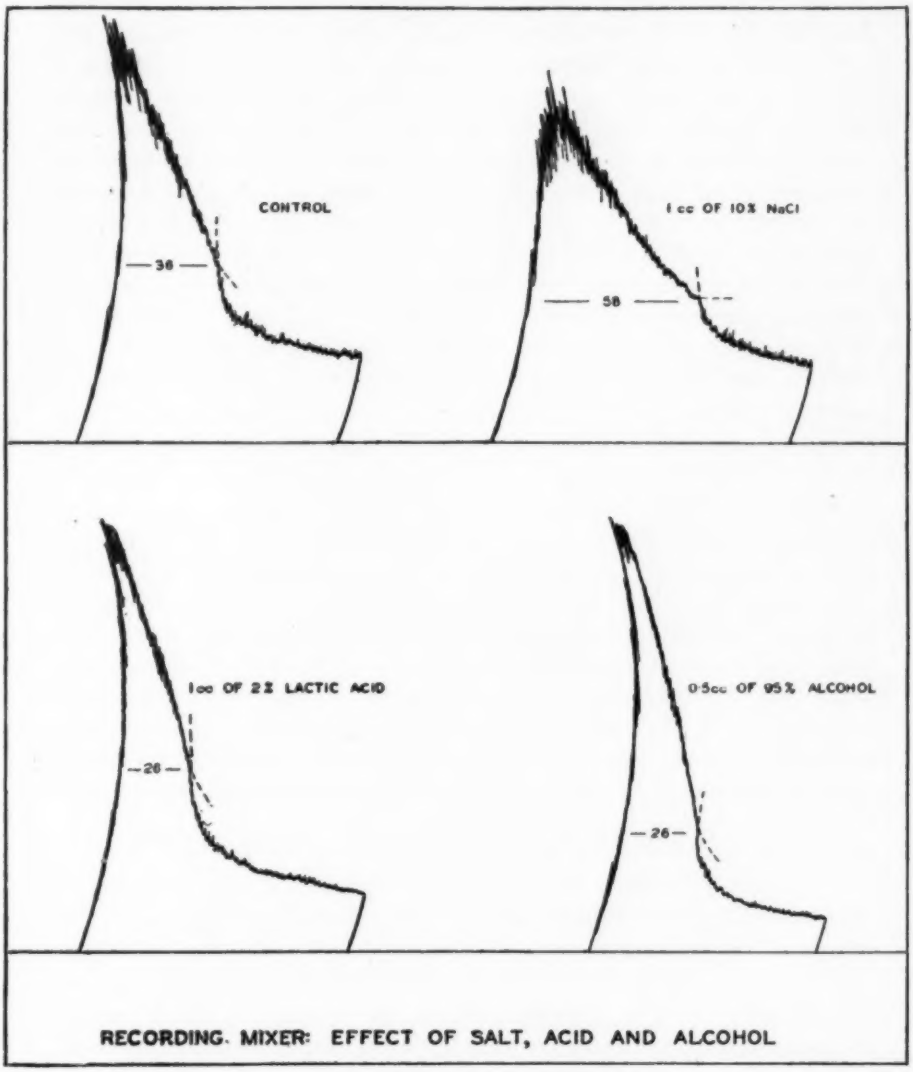

Figure 8. Effect of salt, acid and alcohol.

should reduce it, both decrease the breaking time. One might speculate that the break is indicative of a modification of the linkages of the gluten network, possibly accompanied by a change in the loosely-bound water.

The interpretation of the breaking time in terms of baking quality is a matter of some difficulty. Obviously, if a relation exists it must be through the gluten quality but as this term is generally used it includes not only the quality of the protein as it exists in the flour, but the modification of this quality by other dough ingredients and by factors operative during fermentation. So, even if there were a perfect relation between the breaking time of flour-and-water doughs and the quality of the gluten as present in the original flour, this quality might be so modified during fermentation that there would be no apparent relation between the breaking time and the gluten quality as inferred from the results of baking tests. The problem is also complicated by difficulties in the statistical analysis of pertinent data. Although it has been shown experimentally that protein content and breaking time are independent, it is difficult to obtain a series where they are not correlated, because of the effect of environment on both. This introduces complications in the application of the partial correlation method. Furthermore, protein quality can only be inferred from the results of baking tests and the inference may be greatly in error. Finally, it is unlikely that the breaking time is a complete measure of protein quality and any relation may be obscured by variation in quality due to unmeasured factors.

It is interesting that on a series of 28 samples a significant partial correlation coefficient ($r = -0.44$, 5% point = 0.38) was obtained between breaking time and loaf volume, holding protein content constant. There is some indication that the relation is markedly nonlinear and therefore that it cannot be properly represented by such a coefficient. The evidence points to the existence of a relation between breaking time and gluten quality but this point obviously requires a great deal more investigation before it can be definitely established.

Use of Mixer with Fermenting Doughs, Gluten and Wheat

Fermenting doughs. The mixer operates satisfactorily with doughs of the consistency normally used in test baking, and changes in the dough during fermentation can be detected. For this purpose dough sufficient to provide the desired number of samples is mixed by hand or in a larger mixer. Aliquots are cut off from this bulk and placed in the recording mixer at the desired intervals. To avoid the possibility of the modification during one run affecting the characteristics at later runs, no sample of dough is used twice in the recording mixer. Figure 9 shows typical curves for fermenting dough and incidentally the effect of bromate on the particular flour. It should be noted that the breaking time is not immediately affected by the addition of bromate but differences appear as the fermentation proceeds.

There are striking differences between flours with respect to change in breaking time during fermentation. This is illustrated by the curves in Figure 10 in which the breaking time is plotted against the length of fermentation. It is interesting that these two flours gave very different relative results when tested on this continent by our American methods and when tested in Great Britain.

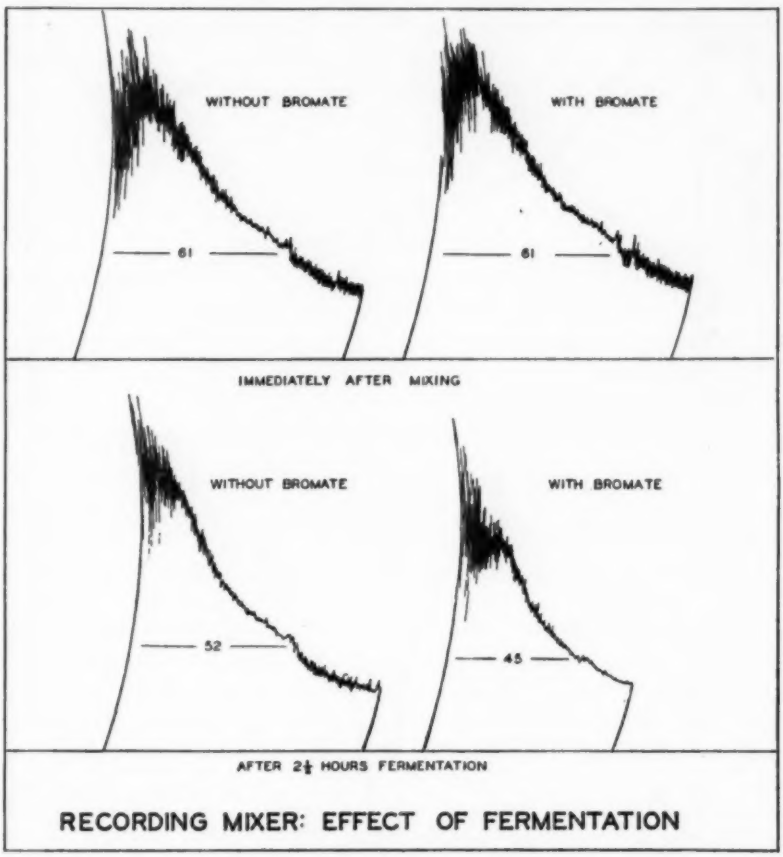

Figure 9. Curves from fermenting dough.

Gluten. The machine gives characteristic curves with both wet gluten and gluten dried by Aitken's method, provided it is suitably diluted with an inert substance such as starch or talc.

Ground wheat. In view of our original objective of providing a method of testing plant breeders' samples, it is of importance that the machine can be used with ground wheat and still more important that there is a close relation between the breaking time of the wheat and of the flour milled from it. In a paired series of 34 samples of wheat

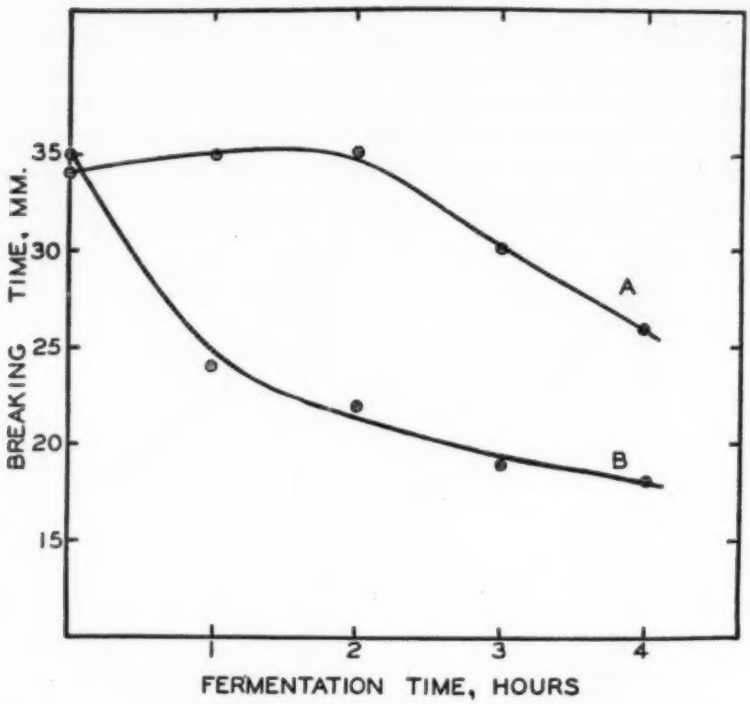

Figure 10. Effect of fermentation on breaking time.

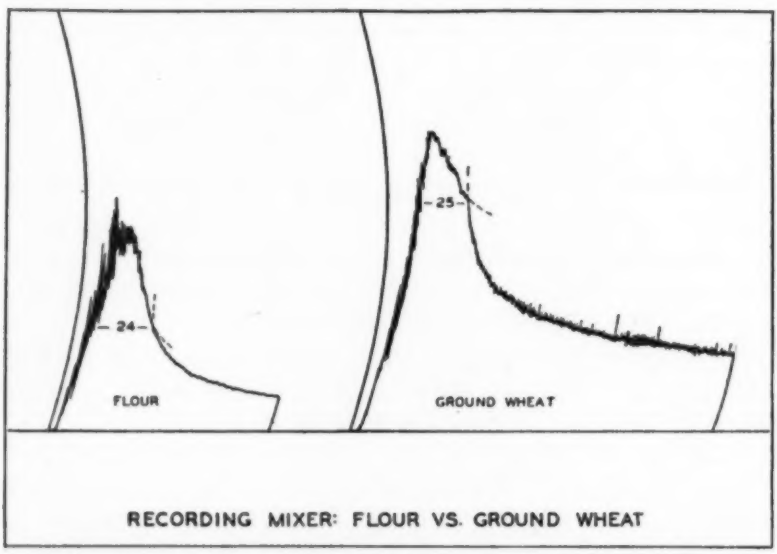

Figure 11. Comparison of curves from wheat and from the flour milled from it.

and 34 samples of flour, the simple correlation between the breaking time of the wheat and the breaking time of the flour was $r = .88$ (1% point = 0.45). Although this relation is not close enough to permit certain prediction, it is close enough for purposes of rough classification. Typical curves of ground wheat and flour from the same sample are shown in Figure 11.

Ground wheat shows the same constancy of breaking time as flour within a normal protein range. However, the protein content below which this constancy is lost is higher with wheat than with flour. With Canadian wheat the change may occur at about 11% (13.5% moisture basis).

Discussion

The experiments made so far have merely opened up the field sufficiently to show that the machine promises to be useful in cereal investigations and to show the lines along which further work may profitably be directed. If the apparent relations between curve characteristics and dough properties can be definitely established, the mixer will be a useful tool in many types of investigation besides the testing of plant breeders' samples, which inspired its construction. However, definite relations must be proven to exist, as the mere fact that flours of different quality give different curves is too general an observation for precise work. The use of such vague terms as dough development to cloak ignorance of the real meaning of curve characteristics is not a satisfying scientific device.

The investigations must proceed along two main lines: (1) attempts to relate curve characteristics to the composition or colloidal properties of the flour or dough, and (2) attempts to relate curve characteristics to baking quality, including dough quality. Neither line of investigation will be easy but a substantial body of information must be gathered if the interpretation of the curves is to rest on a sound foundation.

Summary

A new recording mixer which operates with flour samples containing 7 g. of dry matter is described.

The curves obtained with different flours show marked differences. There is a break in curvature in each curve and this break varies in position and sharpness.

The height and sharpness of the break are both related to the protein content of the sample.

The time at which the break occurs is independent of the protein content, the amount of water added, and the weight of the sample,

but is affected by the addition of reagents known to affect colloidal properties.

There are indications that the time of the break is related to gluten quality.

The mixer can be used to follow certain changes in fermenting doughs.

Curves can be obtained from wet or dried gluten diluted with starch or talc.

The breaking time of curves obtained with ground wheat is closely related to the time obtained from corresponding samples of flour.

Acknowledgment

The author wishes to acknowledge indebtedness to his colleagues on the Associate Committee on Grain Research for supplying him with samples and to his laboratory assistant, K. Hlynka, for his contribution to the constructional details of the mixer.

THE COLLOIDAL BEHAVIOR OF FLOUR DOUGHS. III. STUDIES UPON THE PROPERTIES OF FLOUR-STARCH-WATER SYSTEMS [1]

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(Read at the Annual Meeting, May 1937)

In the preceding sections of this study the properties of starch-water and flour-water systems under agitation in the bowl of a recording dough mixer were investigated (Markley 1937,[2] 1938 [3]). The starch-water system was of a thixotropic nature with the maximum viscosity when at rest but with a decreasing viscosity when in motion. The flour-water systems were quite different; in them the viscosity tended to increase with mixing to a maximum, then to decrease. The principal factor differentiating flour from wheat starch is the gluten. Chemists and bakers have long assigned a role of major importance to the gluten in bread production. In the preceding paper it may be noted in Table I that the low-protein cake flour took less water and less time for development to the point of minimum mobility than did the high-protein spring wheat flours. However, it was not at all clear as to whether this observed difference in flours of diverse protein content was

[1] Paper No. 1523, Scientific Journal Series, Minnesota Agricultural Experiment Station.

[2] Markley, M. C. The colloidal behavior of flour doughs. I. The thixotropic nature of starch-water systems. Cereal Chem. **14**: 434–436 (1937).

[3] Markley, M. C., and Bailey, C. H. The colloidal behavior of flour doughs. II. A study of the effects of varying the flour-water ratio. Cereal Chem. **15**: 317–326 (1938).

due to quantity, or to quality of the protein, or both. The investigation of this point was then attempted.

The effect of varying the quantity of protein while holding quality constant could be studied in either of two ways. One would be to concentrate the protein by a washing out of some starch, but this method has the disadvantage of being unsuited to dough formation studies. The other method is to dilute the flour with wheat starch. While there are some decided objections to this procedure, yet none appeared important enough to make it inadvisable, and accordingly three of the flours used in the preceding section were selected for this purpose. One was the high-protein spring-wheat straight-grade flour, the second the straight-grade flour milled from the semi-hard Minnesota winter wheat, while the third was the low-protein, soft, cake-type patent flour. The starch used in this study was confectioners' grade wheat starch graciously donated by the Huron Milling Company of Harbor Beach, Michigan.

The method of attack upon this problem was to make blends of the flours with the starch in all proportions from 100% flour to 100% starch. Doughs were mixed in the Farinograph from 300 grams of flour at 13.5% moisture with sufficient distilled water to reach a constant minimum mobility of 550 ± 20 Brabender units at 30° C. The resulting curves were measured for time to reach the point of minimum mobility. The data from this study are given in Table I.

The relation between the protein content of the flour-starch blends and the absorption required to bring the doughs to a constant minimum mobility is graphically shown in Figure 1. As the protein content of the blend is reduced from that of the natural flour the absorption tends to decrease, but when the protein has been lowered to about $7\frac{1}{2}$% there appear minima in the absorption, and as the protein content is further lowered below the $7\frac{1}{2}$% level the absorption begins to rise until the absorption of the 100% starch is about the same as that of the high-protein spring-wheat flour.

From these curves it is possible to differentiate between flours as to the relative water-holding powers of their proteins. To illustrate this the spring-wheat flour in its natural state at 14.3% protein required 69% of water to form the standard dough, but at its minimum at 7.3% protein required only 62.5%; while the soft flour of 7.9% protein required 54% in natural state and 53% at its minimum of 7.1% protein. The natural flours had a difference of 15% in their water-absorbing power, but after the flours had been brought to a corresponding protein level by the additions of starch there was still a difference of 9.5% in absorption which is about 2/3 of the original difference. This probably indicates a greater water-binding power of the hard-

TABLE I

FLOUR-STARCH-WATER DOUGHS MIXED IN BRABENDER FARINOGRAPH AT MINIMUM MOBILITY OF 550 BRABENDER UNITS

Flour	Starch	Protein	Absorption	Time to point of minimum mobility
Flour No. 16245, High-protein Spring-wheat Straight				
%	%	%	%	*minutes*
100	0	14.3	69.0	10.50
98.2	1.8	14.0	68.5	9.00
95	5.0	13.6	68.5	13.00
90	10.0	12.9	67.5	8.25
85	15.0	12.2	65.5	7.00
80	20.0	11.5	65.0	6.50
75	25.0	10.8	64.5	5.00
70	30.0	10.1	64.5	5.25
65	35.0	9.4	64.0	3.00
60	40.0	8.7	63.0	1.75
55	45.0	8.0	63.0	1.75
50	50.0	7.3	62.5	1.25
45	55.0	6.5	63.0	0.75
40	60.0	5.8	63.5	1.25
35	65.0	5.1	64.0	1.00
30	70.0	4.4	65.5	1.00
25	75.0	3.7	66.0	1.25
20	80.0	3.0	66.5	1.25
15	85.0	2.3	67.0	1.25
10	90.0	1.6	68.5	1.40
5	95.0	0.8	69.5	1.50
0	100.0	0.2	70.5	0.75
Flour No. 16246, Minnesota Winter Straight				
100	0	12.00	58.5	6.00
95	5	11.40	58.5	5.00
85	15	10.20	58.5	4.50
80	20	9.65	58.0	4.75
75	25	9.05	58.5	1.50
70	30	8.45	57.5	1.75
65	35	7.90	57.5	1.25
60	40	7.30	59.0	1.25
50	50	6.10	60.5	1.25
40	60	4.90	61.0	1.25
30	70	3.75	63.5	1.25
20	80	2.55	66.0	1.25
10	90	1.40	68.5	0.75
0	100	0.20	70.5	0.75
Flour No. 15964, Soft Cake Patent				
100	0	7.90	54.0	1.00
95	5	7.50	53.5	1.70
90	10	7.10	53.0	1.50
85	15	6.75	53.5	1.50
80	20	6.35	54.0	1.00
75	25	5.95	54.4	1.20
70	30	5.60	55.0	1.00
60	40	4.80	56.0	1.00
50	50	4.05	59.0	1.00
40	60	3.25	60.0	1.00
30	70	2.50	63.5	0.70
20	80	1.75	66.0	0.70
10	90	0.81	68.5	0.70
0	100	0.20	70.5	0.75

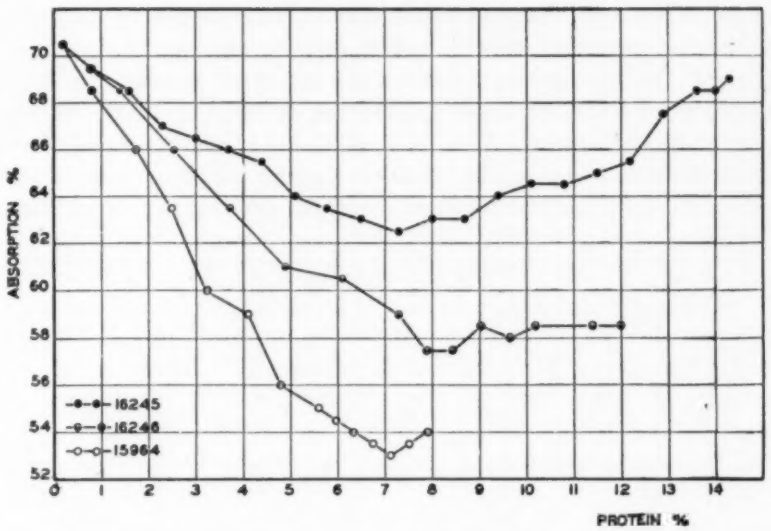

Figure 1. Relation between the protein content of flour-starch blends and the amount of water required to produce doughs of 550 Brabender units minimum mobility.

wheat flour as compared to the soft cake flour. It does not seem quite as probable that the starches involved would be sufficiently different in water-binding power to account for this difference, though such might be the reason.

The relation between the protein content of the flour-starch blends and the time required to bring the doughs to the point of minimum mobility is graphically shown in Figure 2. In this figure it may readily be seen that as the protein content is decreased the time required to develop the dough also decreases simultaneously until a protein level of 8% is reached. At protein levels below 8% the mixing time is constant at about one minute. No differentiation could be noted between the three flours used in this study.

This broken curve is of great interest. The characteristics of the individual Farinograph charts at protein levels below 8% were all very similar to the starch curve presented in Figure 1 of the first section of this study. At levels around 8% the curves from all flours were identical with very short mixing times, typical of the low-protein flours. As the protein content increased the development time likewise increased until the curves were like that in Figure 1 of the second section (Markley, 1938). The break-point in the protein-development time relation occurring at 8% protein corresponds quite well with the 7½% protein minimum for the protein-absorption relation. It would appear that at least 7% of protein must be present if there is to be a continuous gluten structure throughout the dough at mobility levels normal to

bread doughs. At the present time the question of gluten structure in cracker and macaroni doughs, and in cake batters is still unanswered. The more protein present above this minimum amount of 7% the higher the viscosity of the dough, primarily because of the attraction between the protein micelles. The first 7% of protein is apparently held in a much different manner in the system. The increasing water-holding capacity of the system as the protein content is reduced below

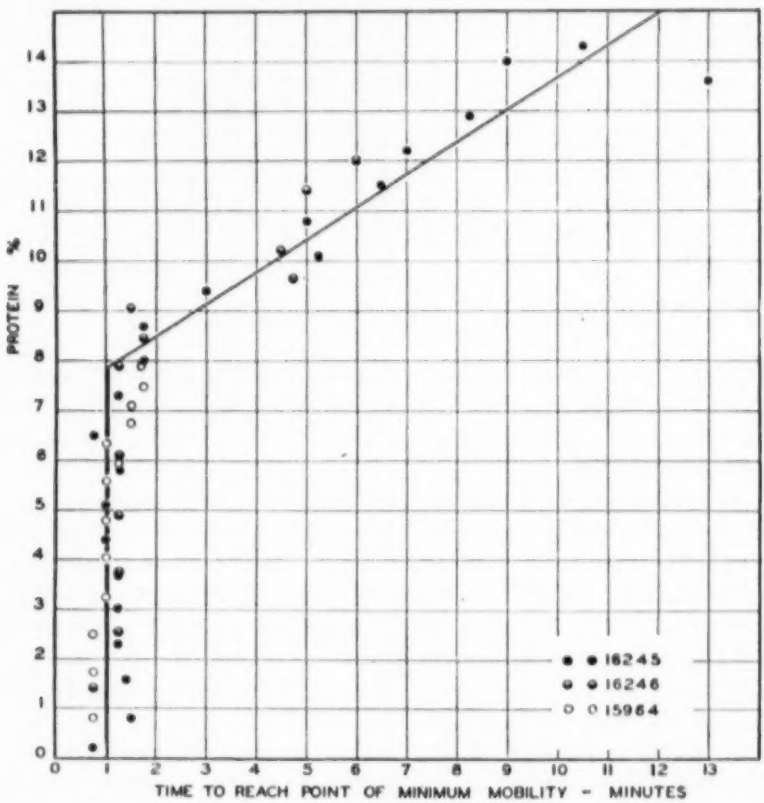

Figure 2. Relation between protein content of flour-starch blends and the time required to mix the doughs to a constant minimum mobility of 550 Brabender units.

7% can possibly be accounted for by postulating an attraction between the starch and the protein which would reduce the water-binding power of both. This may be a form similar to coacervation with all the charges on the protein micelles bound by the starch, and with the protein forming a film over the surface of the starch granules. Possibly 7% protein is just enough to coat the starch, and at this point there is a minimum of attraction between particles or for water. With increas-

ing amounts of protein above 7% there are more active charges upon the proteins than can be held by the starch, and as a result the starch granules are held together by interlocking protein micelles extending out from the protein films over the granules. More water is bound in such a system as well. When the protein content is reduced below 7% there is not enough protein to cover the surface of the starch and the formation of a rigid but fugitive starch gel is possible. This would account for the temporarily higher absorption of the low protein and starch doughs. Such a picture as this needs confirmation by other physical techniques before it is worthy of serious acceptance.

The viscosity of a starch-water paste is primarily a function of the ratio of the volume of the dispersed solid phase to the volume of water. The volume of the solid phase not only includes that of the starch granule itself, but also that of a shell of water more or less tightly bound to the surface of the granule by surface forces of one type or another. This shell can be removed by prolonged shearing stresses as in a dough mixer. The addition of protein up to 7% must reduce the particle size since less water is needed to maintain a constant maximum viscosity in the system. The reduction in particle size by the addition of protein up to the 7% level probably is due to the formation of a more or less complete film or envelope of protein over the surface of the starch granule, with a partial satisfaction of the surface forces by the starch to protein attraction, resulting in a smooth granule with but little water-shell and little attraction to similar particles. If all the starch granules were of a uniform size then the thickness of this film would be about one twenty-fifth of the radius of the granules, but since starch granules vary widely in size this is a rather meaningless dimension. When the starch granule is just covered by the protein film, then the surface forces of the starch must almost exactly balance those of the protein shell, with the result that at this protein-starch ratio the volume of the individual particles is at a minimum. If the granule is not completely covered then there is a partial water shell, held by the unsatisfied surface forces of the starch, with the result that the viscosity is intermediate between that of starch and that of 7%-protein flour. If more than 7% protein is present, then the protein shell would be thicker and more irregular with many exposed secondary valence forces. This would result in both particle to particle attraction and the binding of water to the particles. The viscosity of the dough would then increase in proportion to the amount of protein present above the 7% level.

An examination of bread crumb stained with Millon's reagent indicates that the starch granules are closely packed and are enveloped in a close-fitting envelope of protein. This can only be seen under quite

high magnification of fragments of crumb in natural shape, not crushed under a coverglass. The starch granules are each covered with protein and crowded closely together with the small granules tending to be in the interstices of the large ones. Probably the free water of the dough occupies the remaining space.

Time is required to produce this type of a structure. Evidence of this can be seen in the long time required to mix a high-protein flour dough free of small lumps and irregularities as compared to the rapid appearance of smoothness in low-protein flour doughs. As the mixer blades turn through a well-mixed dough the action is probably not one of tearing apart long strands of gluten, but a pushing of the protein-covered starch granules past each other. These starch granules tend to cohere in proportion to the amount of protein above the 7% minimum in the system. This coherence of the protein-covered starch granules in a flour dough is responsible for the tenacity and ductility of the system, and in part for the viscosity or mobility. The starch granules tend to impart rigidity to the dough. A measure of rigidity is essential in a bread dough. A lack of this property is a cause of the sinking of bread after removal from the pan.

Summary

Absorption of flour-starch-water doughs is at a minimum at approximately 7% protein.

Magnitude of the absorption at 7% protein level for doughs made from mixtures of flour and starch with water is a measure of the water-holding quality of the gluten independent of its concentration.

Doughs made from flour-starch mixtures of less than 7% protein have the physical characteristics of starch pastes.

Doughs made from flour-starch mixtures of more than 7% protein have the physical characteristics of normal flour doughs of similar protein content.

Development time of doughs is a function of the amount of protein over 7%; at levels below 7% there is no differentiation.

The gluten in bread crumb appears to be in the form of an envelope around each starch granule.

About 7% of gluten is required to form the protein envelope around the starch granules.

THE CHEMISTRY OF THE RYE GERM. IV. ITS PROXIMATE COMPOSITION [1, 2]

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(Received for publication January 31, 1938)

A survey of the literature pertinent to the chemistry of the rye germ when evaluated in the light of the volume of similar information which has been published on other cereals, notably wheat, suggests the thought that, in this respect, this field has been neglected. Since rye flour has been for centuries one of the staple foodstuffs of the peoples of Teutonic or Slavic origin, it is but natural that this literature should come almost exclusively from Europe, for there the problem of a wise disposal of the by-products of the milling of rye is of sufficient economic importance to have raised the question of the value of the germ to agriculture in terms of animal feeds.

Inasmuch as existing data on the proximate composition of the germ are not only in a sense incomplete but also not in agreement within tolerances explainable on the basis of differences in cultivation, growth habits, variety, etc., there is an obvious need for new data and a re-evaluation of old. Contributive to these ends, this communication marks the result of attempts to extend the borders of information on the composition of the germ itself beyond that point where they were left by Kling (1910), Rubner (1916), and Kalning (1917) with respect to the organic matter. It records, also, new data on its inorganic constituents and makes comparison, on the one hand, with data recorded in some seven other miscellaneous reports in which the relevant information has been obtained incidental to various and different objectives and, on the other, with those which reveal the composition of the germs of two economically important Gramineae, wheat and corn.

Experimental

Rye germs,[3] recovered from so-called rye-germ stock by repeated screening and air-cleaning, were hand picked for the purpose of removing the small amount of foreign matter which had escaped the first

[1] This investigation was supported by a grant from the Wisconsin Alumni Research Foundation whose aid is gratefully acknowledged.

[2] For previous communications in this series see Stout et al., J. Am. Chem. Soc. **54**: 3298 (1932); **56**: 210 (1934); Schuette and Palmer, Oil and Soap **14**: 295 (1937).

[3] Acknowledgment is made to Frank H. Blodgett, Inc., of Janesville, Wisconsin, who furnished the material used in this investigation.

treatment. Standardized methods of analysis (A. O. A. C.) were used throughout the course of this study. Exceptions were the modified 72% sulfuric acid method of Peterson *et al.* (1932) for lignin and the phytin phosphorus procedure of Averill and King (1926). Sucrose was determined from its rotation (9.20%) and from the yield of Cu_2O after hydrolysis (9.33%). The average of the values so obtained is reported. For the determination of copper the Elvehjem-Lindow (1929) modified pyridine-thiocyanate method was used. Lipoid phosphorus represents that which is present in the ether extract or crude fat.

Results

The composition of the germ with respect to its organic constituents (see Table I) is reported on a moisture-free basis, the calculation resting upon a loss of 7.86% on heating under reduced pressure at 70°.

TABLE I
ORGANIC CONSTITUENTS OF RYE GERM

	%	%
Ether extract		13.23
Crude protein (N × 5.83)		39.76
Nitrogen of alcohol-soluble proteins	0.50	
Albuminoid nitrogen	6.06	
Nitrogen not accounted for	0.25	
Carbohydrates		27.37
Sucrose	9.28	
Raffinose	2.15	
Starch (by diastase)	7.18	
Pentosans	8.76	
Lignin		6.82
Crude fiber		2.44

Proteins soluble in relatively strong alcohol (70%) constitute 7.3% of the whole. Similarly, for albuminoids this factor is 89. The organic bases probably account for the remainder, a situation already known to obtain, according to Power (1913), with at least one other of the Gramineae.

A total phosphorus content of 0.66% was found, of which approximately 18% has been accounted for in organic combination, *viz.*: phosphatides as lecithin 0.364% and phytin 0.39% (P × 3.55).

The so-called "nitrogen-free extract" amounts to only 5.4%, which value is reduced by 7% of itself if the figure found for cellulose (0.38%) be deducted. But, because of the unsatisfactory state of the pertinent analytical procedure, some doubt exists as to the validity of the latter in spite of the fact that excellent check results were obtained. Be that as it may, however, the low order of magnitude of the former value quite properly may be construed as an indication that the analysis has been fairly complete within the scope of the limitations of investigations of this type.

The ash content of the moisture-free germ was found to be 4.97%, with water-insoluble constituents (52.7%) predominating over the water-soluble. The alkalinity of the latter, however, exceeds by a small amount that of the former, or 1.20 and 1.04 c.c. 0.1 *N* acid per gram of sample, respectively. The content of each inorganic constituent, calculated to the form indicated within the accompanying parenthesis (see Table II), is expressed both in relation to the original germ and the total ash and, for the purpose of determining the potential acid-base balance of the germ along the lines followed by Sherman and Gettler (1912), in terms of the equivalent volume of normal acid or base solution per 100 g. of substance.

TABLE II

INORGANIC CONSTITUENTS OF RYE GERM

Constituent	Concentration		Acid or base equivalent N/1 sol. per 100 g.
	% of germ	% of ash	*C.c.*
Iron (Fe_2O_3)	0.033	5.63	3.4
Aluminum	trace	—	—
Manganese (Mn_3O_4)	0.024	0.48	0.6
Calcium (CaO)	0.090	1.81	3.2
Magnesium (MgO)	0.530	10.66	26.6
Sodium (Na_2O)	0.130	2.61	4.0
Potassium (K_2O)	0.510	10.26	10.8
Copper (Cu)	0.00056	0.01	—
Zinc (Zn)	0.0166	0.33	—
Silica (SiO_2)	0.052	1.04	3.4
Phosphorus (P_2O_5)	1.511	30.40	44.0
Sulfur (SO_3)	1.075	21.62	27.0
Chlorine (Cl)	0.085	1.71	2.4

Discussion

In so far as the results disclosed by the three major literature citations in question (see Table III)—they have been taken from German sources and presumably have reference to rye produced in that country—may serve as the yardstick for evaluating an American-grown product, it appears that there are in this instance no very significant differences in respect to content of either total oxidizable organic nutrients or of inorganic constituents. Specifically, the observed crude fat content of 13.23% lies within the reported limits of 11.95 and 14.44%; it is practically the same as the calculated average for this group but higher than that (10.71%) of all like data heretofore reported by others. The crude protein value of 39.76%, although substantially the same as the all-average of 40.36%, is below the 41.05-minimum of Rubner (1916), itself less than the maximum content, or 46.31%, which Kling (1910) found.

TABLE III

COMPARATIVE DATA ON THE COMPOSITION OF RYE GERM

Constituent	Kling-Rubner-Kalning		All available		Wisconsin, %
	Limits, %	Average, %	Limits, %	Average, %	
Crude fat	11.95–14.44	12.93	7.30–14.44	10.71	13.23
Crude protein	41.05–46.31	44.03	25.85–44.74	40.36	39.76
Pentosans	7.32– 8.04	7.56	7.32– 8.04	7.56	8.76
Crude fiber	2.63– 3.94	3.28	1.83– 6.70	4.41	2.44
Nitrogen-free extract	32.81–33.83	33.32	28.65–51.43	37.64	30.84
Ash	5.54– 6.76	6.05	4.43– 9.65	6.43	4.97

Almost three decades ago the first information on the pentosan content of the germ (8.04%) was announced. Credit for this belongs to Kling (1910). Within short intervals thereafter appeared the practically identical data (7.32%) of Rubner (1916) and Kalning (1917), the average value for the group then becoming 7.56%, one which is about one-seventh less than that found for the domestic product which is 8.76%. Kalning (1917), apparently with indifferent success, alone appears to have attempted a determination of the starch content of the germ. He reported a value of *ca* 6%, one which at least approximates the order of magnitude of that herein recorded, or 7.18%. These findings, therefore, seem to invalidate the statement of Kling (1910) that rye germ contains no starch.

Because of the nature of the determination itself, one leading to results which, in a sense, are defined by the technique followed, it is perhaps to be expected that wide variations in the crude fiber content of an agricultural product will be reported. The rye germ is no exception for witness the minimum of 1.83%, a 3.7-fold maximum and an average —nine reports are involved—of 4.41%. Kling's (1910) value of 2.63% alone in this group compares with that reported herein, or 2.44%. The cellulose content of 3.13% which Rubner (1916) reported might well be accepted with some reservations for reasons above noted. It is eight times larger than that herein reported.

Sugar determinations have been attempted by Kalning (1917) but, unfortunately, his results, which were reported as reducing sugars before hydrolysis 6.66% and sugar after inversion 22.62%, are not very informative. Kling (1910) dismissed this phase of his investigation with the observation that the nitrogen-free extract (32.81%) may be deemed to consist practically all of carbohydrates. Frankfurt (1896) indicated that the sucrose, raffinose and glucose content of wheat germ totals 24.34% and later Power and Salway (1913) isolated the first two

from this source. Our own investigations have shown that rye germ contains 9.28% sucrose and 2.15% raffinose.

The hitherto published lower and upper limits of the mineral matter of this germ, as revealed in eleven communications, are 4.43 and 9.65% —the average of them all is 6.43%; for those reported by the three investigators cited it is 6.05%. The majority of the indicated values, however, are of the order of magnitude (4.97%) herein reported.

If anyone before has quantitatively examined the nature of the inorganic constituents of the rye germ, then this fact has escaped notice in a meticulous search of the literature on the subject. The results of such an analysis calculated to a dry basis are presented in Table II. They reveal the fact that rye germ is rich in phosphorus (less so in this respect than is wheat germ), sulfur, magnesium and potassium. Whether the apparently high content of iron and magnesium might not be in part due to an incidental contamination in the milling process is a matter for conjecture. The copper content of the germ is approximately that of white bread, Graham flour, dried dates and figs, oatmeal, sweet potatoes, and sweet Bantam corn (Lindow *et al.*, 1929) but less than that of the wheat germ which Elvehjem and Hart (1929) found to contain 9 mg. per kilogram. An interpretation of these data from the standpoint of potential acid-base balance by the Sherman-Gettler (1912) calculation reveals a preponderance of acid-forming elements. Although it appears that phosphorus (0.66%) is the most prominent element in this group, yet its concentration in the wheat germ is apparently greater. Andrews and Bailey (1932) found that the phosphorus content of wheat germ is 1.244%, that that in phytic combination constitutes 47.98% of the whole, and that in lipoid form 5.70. Comparable data for rye germ were found to be 16.6 and 22.7% respectively.

There remains to be made, finally, comparison of the proximate composition of rye germ as herein reported with similar data on wheat and corn germ as gleaned from the literature and collated in Table IV.

TABLE IV

PROXIMATE COMPOSITION OF THE GERMS OF THE GRAMINEAE

	Wheat	Corn	Rye
Protein	23.60–40.75[21] [1]	11.98–15.50[4]	25.85–44.74[11]
Fat	6.01–13.51[22]	3.72–24.36[4]	7.30–14.44[11]
Crude fiber	1.63– 4.75[16]	4.98– 5.76[3]	1.83– 6.70[9]
Starch	9.95–13.50[3]	24.75–47.50[2]	6.00[1]
Pentosans	10.92[1]	—	7.32– 8.04[3]
Ash	3.90– 6.40[20]	2.77– 6.05[4]	4.43– 9.65[11]
Nitrogen-free extract	39.25–48.50[14]	45.42–48.72[3]	28.65–51.43[10]

[1] Superior numbers indicate the number of pertinent samples.

This compilation is presented without literature citation for reasons of economy of space. It should be read with reference to the last column in Table III.

Summary

Rye germ is a high-protein, phosphorus-rich substance in which the lipoid predominate over the phytin forms of the latter. In Germany it has received some measure of study as to its suitability for animal feeds. The opinion there expressed is that farm animals relish it and assimilate it readily. Incidental to such investigations there have been made studies in European laboratories on composition which, although doubtless extensive enough to serve the immediate objectives then in hand, admit of elaboration. A study of this character has been made on an American-grown product. No very significant differences in respect to content of either total oxidizable organic nutrients or inorganic constituents have been found. The so-called nitrogen-free extract has been broken down to the point where there remains *ca* 5% to be accounted for. A quantitative examination of the ash has shown that acid-forming elements predominate over the base-formers.

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A STUDY OF THE BACTERIAL POPULATION OF GRAINS USED IN A DISTILLERY

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(Received for publication March 17, 1938)

Introduction

In preparing grain mash in a distillery, effective sterilization by heat is impossible both for economical and operative reasons. Corn is usually pressure-cooked but rye is normally mashed at 160° F. Malting or starch conversion operations are carried out at temperatures below 160° F. so as not to impair the desired enzymatic activity of the malt. Therefore, if a low bacterial count of the mash is to be accomplished, a careful check of the grains used must be made and those grains showing a high count must be rejected.

There are two objections to a high bacterial count in the mash. First, even a moderate number of bacteria in the original mash means there will be a much larger number present after one or two days of fermentation at 75° F. to 90° F. (24° C. to 32° C.). These organisms will grow at the expense of some of the carbohydrate present, thereby lowering the yield of alcohol. Second, while the yield of alcohol is being lowered, objectionable bacterial fermentation products are being formed and these adversely affect the quality of the distillate. Those by-products which are steam distilled with alcohol, and hence become a part of the whiskey, are especially objectionable.

Methods

Hoffman, Schweitzer, and Dalby (1937) have contributed a method of counting spores responsible for "rope" in bread, but their technique is not suited for the determination of total bacterial counts in grains.

Brahm (1921) contributed studies on the bacterial flora of flour and was probably the first investigator to propose bacteriological standards for this commodity. He proposed a limit of 150 "rope" spores per gram of flour for bakery use.

Since there is no standard or official method for the estimation of numbers of bacteria in grain, it was necessary to work out a practical method that would combine accuracy, speed, and ease of operation.

In determining the bacterial count of grains, important points to be observed are proper sampling, the avoidance of contamination in

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grinding and handling, the sterility of all glassware, diluting water and media, proper incubation, and lastly, proper counting.

Sampling. A probe sampler is used which collects 10 individual samples at one time. Five thrusts are made with this sampler at different points in the car of grain and the fifty individual samples composited in a clean, dry container. This is the standard procedure used to collect samples for moisture, damage, and other standard and special grain tests. After thorough mixing, the large sample is reduced by halving until about 50 g. remain.

Grinding. The 50-g. sample is next ground to a fine flour in a clean, dry mill. A No. 1 Wiley Mill using the 1 mm. screen is used for this purpose.

Numerous screen passage determinations have shown that this grinding gives a meal fine enough so that 95 to 97½% will pass a 30-mesh screen, with less than 1% being retained on a 20-mesh screen.

Sterilization of glassware and media. All glassware and media are prepared and sterilized in accordance with the recommendations of "Standard Methods for the Examination of Water and Sewage" of the American Public Health Association (1928). Difco Nutrient Agar is the medium used for plating.

Water blanks. In the Hiram Walker laboratory an effort has been made to reduce diluting transfers to a minimum, and for this reason, only 99 c.c. water blanks are used. These are prepared by filling 250 c.c. capacity round bottles with approximately 103 c.c. of water so that 99 c.c. remain after sterilization.

Rubber stoppers containing two inches of glass rodding are used instead of cotton plugs. The glass rod acts as a mechanical agitator during shaking and extends into the bottle when the stopper is in place.

After the dilution bottles have been filled with the required amount of water they are packed loosely in the autoclave without the stoppers in place. The stoppers are either placed on end or in a wire basket and sterilized along with the dilution water. Sterilization is for 20 minutes at 15 pounds pressure.

After the pressure in the autoclave has been released, the stoppers are immediately inserted into the water blank bottles. This operation is carried out within the autoclave. After the bottles are removed from the autoclave, they are allowed to cool before use.

Plating. Two dilutions can normally be run, namely, 1 to 100, and 1 to 10,000. One gram of the ground grain meal is weighed out on a balance and introduced into a 99 c.c. water blank. The stopper is not touched where it will come in contact with the inside of the bottle. During the introduction of the meal, the stopper is rested on the desk with the glass rod straight up, and after the introduction of the meal it is immediately replaced.

The bottle is then shaken vigorously for 20 seconds timed with a watch to obtain an even suspension of the finely ground meal and micro-organisms. If the grain is known to be of a low bacterial count, this 1 to 100 dilution is all that need be made.

The next higher dilution is made by withdrawing one cubic centimeter from the first dilution with a sterile pipette and introducing it into a second 99 c.c. water blank. This 1 to 10,000 dilution is sufficiently high for most grains. This second dilution bottle, like the first, is then shaken for 20 seconds in order to obtain an even suspension of organisms.

Duplicate petri-plates are prepared from the dilution selected by withdrawing one cubic centimeter of the suspension and introducing it in the center of the plate by means of a sterile pipette. Melted nutrient agar is next added, and with a swirling motion, the sample and agar mixed, the plate covered, and then left to harden. Incubation is for 24 hours at $37\frac{1}{2}°$ C.

The counting of the plate after incubation is done in the usual way with the aid of an illuminated counting apparatus.

Accuracy of the method. The accuracy of any method cannot be proved until a number of workers in different laboratories have compared results of the method. The comparisons that have been made to date on this method indicate results that check within 10% (plus or minus) in grains containing approximately three million bacteria per gram.

Grain Counts

Bacterial count studies have been made of all grains used in the distillery. In Table I there are listed the monthly averages of the four

TABLE I

MONTHLY AVERAGES OF GRAIN COUNTS, OCTOBER 1936 TO MAY 1937

Total Bacteria Per Gram

Date	Barley malt	Rye malt	Rye	Corn
1936 October	2,400,000	———	800,000	15,000
November	2,800,000	1,000,000	900,000	35,000
December	2,700,000	3,600,000	1,200,000	2,400,000
1937 January	5,200,000	3,800,000	1,000,000	2,000,000
February	4,500,000	———	1,400,000	300,000
March	3,800,000	3,900,000	1,700,000	3,500,000
April	2,800,000	3,300,000	1,100,000	1,500,000
May	3,400,000	1,900,000	1,600,000	900,000
June	3,900,000	2,600,000	800,000	200,000

grains used from October 1936 to May 1937. Table II lists the maximum, minimum, and average count data of the four grains for the month of June 1937.

TABLE II

MAXIMUM, MINIMUM, AND AVERAGE COUNTS OF GRAINS SAMPLED IN JUNE 1937

Total Bacterial Count Per Gram

	Maximum count	Minimum count	Average count for month
Barley malt	7,700,000	700,000	3,900,000
Rye malt	4,600,000	1,600,000	2,600,000
Rye	1,700,000	60,000	770,000
Corn	800,000	30,000	240,000

From these tables it will be seen that of the four grains used, corn has the widest variation in its bacterial count. During October and November 1936, corn of the 1935 harvest was being used which had an average moisture content of only 13% and low bacterial count. During December a mixture of new-crop kiln-dried and new-crop natural-dried corn was used. It is noted that this corn showed a much higher average bacterial count. In February only kiln-dried corn of a low count was used. During March, April, and May, natural-dried corn of the 1936 crop was used. The moisture content of this corn averaged 15.9, 17.1, and 15.8%, respectively, for the three months. Increased bacterial counts are noted for this natural-dried corn over the kiln-dried corn used exclusively during February. This wide fluctuation in bacterial count is due to the type of drying the corn receives and its final moisture content, as well as the number of damaged kernels present.

Studies of the bacterial count of kiln- and natural-dried corn have been made and the averages of several determinations are given in Table III.

TABLE III

EFFECT OF KILN DRYING ON BACTERIAL COUNTS OF CORN

Type of drying	Number of samples	Bacterial count per gram	Moisture %
Natural dried	20	3,725,000	17.3
Kiln dried	12	990,000	14.5

The heat treatment necessary for kiln drying not only reduces the moisture content of the corn but markedly reduces the bacterial count. In the samples shown in Table III, this reduction in count amounts to about 400%.

Samples of damaged and undamaged corn kernels have been assayed for bacterial population. The results show that counts on damaged kernels run as much as 285 times greater than those of the

undamaged kernels. The data from typical studies are given in Table IV.

TABLE IV

EFFECT OF DAMAGED KERNELS ON BACTERIAL COUNTS OF CORN

Material	Bacterial count per gram	Ratio of counts on damaged and damage-free kernels
Damaged kernels	18,560,000	1 to 285
Damage-free kernels	40,000	
Damaged kernels	6,464,000	1 to 139
Damage-free kernels	45,000	

The malts have by far the highest bacterial count of the four grains used in a distillery. This is not surprising when one considers the nature of their preparation, for at no time after steeping and germination do they receive a heat treatment above 125° F. (52° C.). Studies in this laboratory have indicated that relatively few organisms are to be found on the outside of the kernels and that the bulk of the bacteria is to be found immediately under the outer covering of the kernel. Private communications from another laboratory have confirmed this finding.

As a result of studies made on the bacterial count of grains and the effect these bacteria have on yields and quality of the final distillate, bacteriological standards have been adopted for all grains used in the Hiram Walker Distillery. The maximum acceptable counts are given in Table V.

TABLE V

MAXIMUM ACCEPTABLE BACTERIAL COUNTS PER GRAM

Material	Maximum acceptable bacterial count per gram
Barley malt	10,000,000
Rye malt	10,000,000
Rye	3,000,000
Corn	3,000,000

It is interesting to note the reductions in bacterial counts that have followed these efforts. Before this work was started, counts as high as 30 to 50 million per gram were commonly found in barley malt. When these results were brought to the attention of maltsters, many cooperated fully in efforts to reduce the count in their products, with the result that malt is now regularly supplied by some maltsters having a count of only one tenth, or even less, than was formerly the case.

Summary

A method for counting bacteria in grain has been developed which is accurate, fast, and simple. Other laboratories using this method have obtained satisfactory checks.

The method is quite flexible, being applicable to grains of all sorts, husks of grains, flour, and related materials.

The development of the method has permitted studies of the bacterial population of grains and its effect on yields and quality of final distillates. These have resulted in the establishment of bacteriological standards for all grains used.

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COMPARATIVE RATES OF AMYLASE ACTION ON STARCHES [1]

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(Received for publication March 17, 1938)

Several investigators (Stone, 1896; O'Sullivan, 1904; Ford, 1904; Nagao, 1911; Sherman, Walker, and Caldwell, 1919; Hermano and Rask, 1926) have reported differences in the rates of amylase action on starches, depending on the origin of the starch. Glock (1936) points out that the results are contradictory and suggests that each investigator may have been measuring a different effect of amylase action. The majority of investigators report potato starch to be more rapidly digested than the cereal starches. In this laboratory, while attempting to ascertain the lowest possible temperature for gelatinization of potato starch pastes for enzyme action, it was found that when the starch was heated at 70° C. for 30 minutes hydrolysis by soybean amylase was much faster than when the starch was boiled. This suggested that the discrepancies in the literature might be due to differences in methods of preparing the starch substrates. Day (1908) studied the effect of cooking different starches on their digestibility by amylase. Potato and arrowroot starch pastes were found to be

[1] Journal Paper No. J543 of the Iowa Agricultural Experiment Station, Ames, Iowa. Project No. 517.

digested as well if made with hot water and not boiled as those which had been boiled three hours. Corn and wheat starches were made somewhat more digestible by long cooking.

The methods and data reported here are a part of a detailed study of the effect of temperature of gelatinization on rates of hydrolysis of five different starches by soybean amylase. The starches used were corn, potato, rice, wheat and tapioca. Substrates were made from these starches by heating at 60°, 70°, 80°, 90°, 100° and 120° C. The rate of hydrolysis of the starch substrates by soybean amylase was measured by a potentiometric determination of the reducing sugars formed.

Preparation of the Starch Substrates

All substrates contained 2% starch and were at pH 5 as measured by the glass electrode. Ten grams of the untreated starch were stirred with 100 c.c. of cold water. This paste was poured into 350 c.c. of a solution containing distilled water and 49.0 c.c. of 0.2M NaH_2PO_4 solution and 1.0 c.c. of 0.2M Na_2HPO_4, which had been brought to the desired temperature in a water bath. The mixture was kept at the desired temperature 30 minutes. It was then cooled to 40° C. and made up to 500 c.c. The substrates heated at 120° C. were prepared by boiling the mixture prepared as the others and then heating under 15 pounds of steam pressure in an autoclave for 30 minutes. The substrate was divided into two 250 c.c. portions and the same amount of enzyme added to each. The digestions were carried out at 40°. Five cubic centimeter portions were removed from each digestion at the same time for the sugar determinations, giving two values for every point on the curves.

Preparation and Activity of the Enzyme

The enzyme used was prepared from ether-extracted soybean meal by extraction with 50% alcohol and precipitation by adding absolute alcohol to make the concentration 70%. The dried powder had a saccharogenic power of 825 and zero dextrinogenic activity (see Ohlsson and Edfeldt, 1933). For each 250 c.c. of the 2% starch, 20 c.c. of a suspension in water containing 40 mg. of enzyme per 100 c.c. were used.

The ratio of enzyme to starch was one to 625, and the ratio of enzyme to maltose at 70% digestion was about one to 400. There was an excess of enzyme present at all times during the digestions.

Determination of Reducing Power of the Digestion Mixtures

It was not possible to use the modified Hagedorn and Jensen method for determining reducing sugars (see Blish and Sandstedt,

1933; Gore and Steele, 1935) to follow the course of these digestions. In the cases where the starch was heated at 60°, for instance, there were particles of unswollen starch in the digestion. These particles adsorbed the iodine and held it so tenaciously that it was impossible to duplicate results. The method of Hassid (1937) was modified to a macro determination, and the titration was followed potentiometrically.[2]

Five cubic centimeters of sample containing from 5 to 60 mg. of maltose were pipetted into 25 c.c. of alkaline ferricyanide reagent.[3] This mixture was placed in a boiling water bath for 15 minutes, then cooled in running water 2 or 3 minutes. Twenty-five cubic centimeters of a 1–4 solution of HCl were added immediately, and the contents of the flask poured into a 250 c.c. beaker for titrating. The flask was rinsed into the beaker with two 10 c.c. portions of distilled water. The final volume of the solution was 80–85 c.c., and the acid concentration 1.0–1.5N. According to Furman and Evans (1929) this is the optimum acid concentration for the following reaction to proceed rapidly and quantitatively.

$$Ce^{+4} + Fe(CN)^{-4} \rightarrow Ce^{+3} + Fe(CN)^{-3}.$$

The solution was titrated potentiometrically with 0.1N ceric sulfate[3] solution (see Furman and Evans, 1929) using a platinum-saturated calomel electrode system and a KCl-agar bridge. These electrodes were found to give an increase of 350 millivolts at the end-point in 1–1.5N acid. This voltage jump was much greater than could be obtained with a platinum-tungsten bimetallic electrode (see Furman and Wilson, 1928). Before titrating the solutions were green, and the color changed abruptly to yellow about 0.05 c.c. before the voltage change.

The results of these determinations were calculated to milligrams of maltose by converting the titration value to cubic centimeters of 0.1N ceric sulfate and reading the value for maltose directly from a graph prepared from data obtained by titrating solutions of known concentrations of C.P. maltose hydrate. The purity of the maltose was checked by the standard Munsen-Walker method. The results of the sugar determinations run on portions of the simultaneous duplicate digestions checked within 0.5 mg. The results on a repetition of the experiment showed a variation which was never more than

[2] The voltages were measured on an experimental model of an instrument operating directly off the 110 volt A.C. line, which can also be used to measure pH with glass or quinhydrone electrodes. The instrument is being manufactured for sale by Precision Scientific Co., Chicago, Illinois.

[3] The reagents were prepared as follows: alkaline ferricyanide reagent—0.1N potassium ferrocyanide (by weighing) in 5% sodium carbonate solution; 0.1N ceric sulfate—53 g. of C.P. ceric sulfate (1.6 times the theoretical) were added to 900 c.c. of a solution containing 100 c.c. of concentrated sulfuric acid. This was digested on a hot plate until all the solid had dissolved. It was then filtered and made up to one liter. The solution was 1.0N in sulfuric acid. The ceric sulfate solution was standardized potentiometrically against a standard ferrous iron solution.

1% on a single determination. The time-maltose curves could be duplicated, therefore, with occasional points about 1% off the curve.

Data and Discussion of the Results

There was very little increase in the reducing power during the digestion of the various starches that had been heated at 60° for

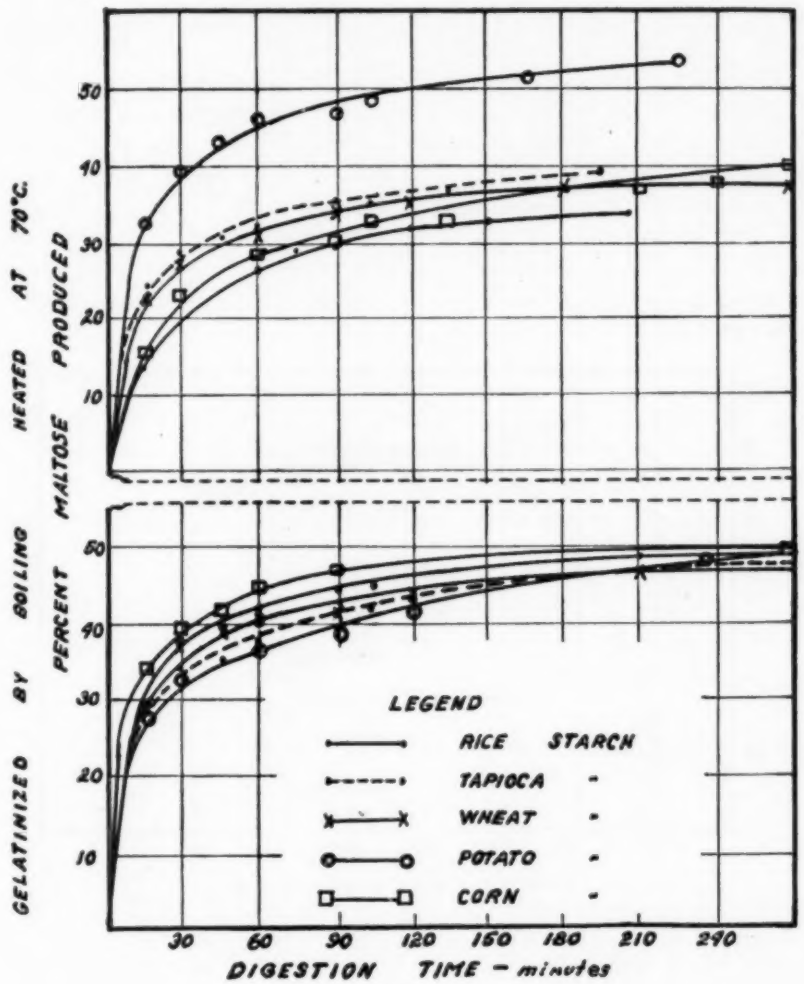

Figure 1.

30 minutes, except in the case of potato starch. Soybean amylase does not digest potato starch heated at 50°. Figure 1 shows the results of the sugar determinations plotted against time of amylase

action for starches which had been heated at 70° and 100° as previously described.

When the starches were prepared by heating at 70° C. for 30 minutes, the potato starch is digested most rapidly by soybean amylase. As shown in Figure 1, the tapioca starch is next in order and wheat, corn and rice are slower than either potato or tapioca. The limit of hydrolysis after 24 hours' digestion of the potato and tapioca starches heated at 70° was between 66 and 68% of the oven-dry starch. The limit for the cereal starches heated at 70° was 49 to 50%.

When the substrates were prepared by heating the starches at 100° for 30 minutes the order of rapidity of enzyme digestion is reversed. Corn starch was digested most rapidly, with wheat, rice, potato, and tapioca less rapidly, in order. The limit of digestion of wheat, rice, and tapioca starches heated at 100° was 57 to 60%, for corn starch 70% and potato starch 65% after 24-hour digestion. Heating the starches at a higher temperature slowed the enzyme action on potato and tapioca starches so much that the cereal starches were hydrolyzed more rapidly.

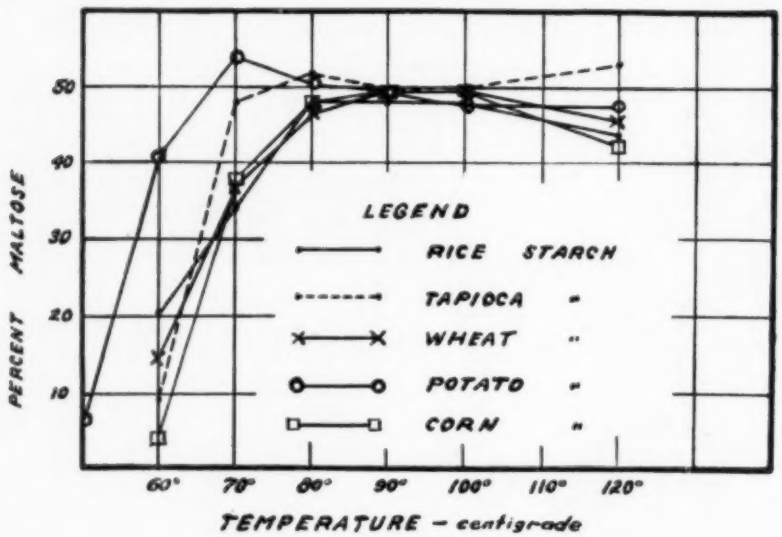

Figure 2.

The effect of heating the starches on the amylase action is shown more clearly by Figure 2 in which the maltose formed in 240 minutes of enzyme action is plotted against the temperature at which the starches were prepared. In every case except rice starch an optimum temperature of preparing the starch for soybean amylase action is indicated—70° for potato, 80° for tapioca, and 90° for wheat and

corn starches. Temperatures of 80° or above for preparing rice starch substrates will give the maximum rate of soybean enzyme hydrolysis.

The decrease in the rate of soybean amylase action on corn, wheat, and potato starches which have been heated above the optimum temperatures is an interesting phenomenon. However, after 24 hours' digestion the different starches prepared at these higher temperatures approach about the same limits of maltose formed as the same starches heated at their optimum temperatures. These results suggest that there is some effect on the starches when heated, other than the swelling and rupture of the granules. It is possible that heating causes agglutination of the particles in the gelatinized starch paste, so that a change in the degree of dispersion occurs. The individual starch molecules would then be less accessible to the attack of the soybean amylase. The result would be a retarding effect, but eventually the same degree of hydrolysis would be accomplished.

Summary

A potentiometric method for determining reducing sugars quantitatively which is applicable to the determination of maltose formed during enzymic hydrolysis of starches has been described.

The temperature at which the starch substrate is prepared has a definite effect on the rate of soybean amylase action.

The optimum temperature of preparing starches for soybean amylase action depends on the kind of starch to be used. The following temperatures of gelatinization were found to give the maximum rates of soybean amylase action on the different starches studied—70° for potato, 80° for tapioca, 90° for wheat and corn starches, and 80° or above for rice starch.

A possible explanation of the retarding effect of heating above these optimum temperatures on soybean amylase action on corn, potato, and wheat starches is given.

Acknowledgment

The authors wish to thank Nellie M. Naylor and R. M. Hixon for their advice and suggestions during the course of this work.

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SOME PHYSICAL FACTORS AFFECTING THE DISPERSION OF SOYBEAN PROTEINS IN WATER

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(Received for publication January 14, 1938)

It is the object of this paper to consider some of the factors which might influence the amount of nitrogenous constituents dispersed in water from fat-free soybean meal and to point out analytical precautions that should be taken when the measurement of this dispersion is attempted.

Considerable data have appeared in the literature (see Bailey, Capen, and LeClerc, 1935; Osborne and Campbell, 1898; and Staker and Gortner, 1931) giving values for the solubility of seed proteins. In most of this previous work the main objective has been to isolate protein fractions and but little consideration has been given to the physical factors which might affect the extraction. A few investigators have realized the significance of these conditions and have published important observations on them. Johns and Jones (1916) noted that there was no appreciable difference in the amount of protein extracted from peanut meal by sodium chloride solutions at 40 to 50° C. and at room temperature. On the other hand O'Hara and Saunders (1937) show that variations in temperature affect the amount of protein extracted from orange-seed meal by 4 N. sodium chloride solution. They also present data to show that within reasonable limits, time is not an important factor in the extraction of flaxseed proteins. Bishop (1929) observed that about 10% more protein was extracted from barley when it had been finely ground. Hofman-Bang (1930) also working with barley states that in order to obtain uniform results the meal should be ground in a ball mill until 95% passes through a 100-mesh sieve. Staker and Gortner (1931) also realized the importance of fine grinding and a standard particle size. They ground the meal of various seeds to pass through a 100-mesh sieve before extraction, but presented no data to justify this choice.

In the work reported in this paper the amount of nitrogenous material dispersed was measured by a Kjeldahl determination on the suspensoid. It has been generally assumed by investigators in this

[1] A cooperative organization participated in by the Bureaus of Chemistry and Soils and Plant Industry of the United States Department of Agriculture, and the Agricultural Experiment Stations of the North Central States of Illinois, Indiana, Iowa, Kansas, Michigan, Minnesota, Missouri, Nebraska, North Dakota, Ohio, South Dakota, and Wisconsin.

field that by far the greater part of the nitrogen exists in the form of proteins. Winton (1935) presented a compilation of the work of three groups of investigators who reported the nitrogen distribution of soybeans. Their work indicates that approximately 5 to 10% of the nitrogen is non-protein.

In a preliminary survey samples of a number of varieties grown in widely separated localities were extracted, but it was soon discovered that variations in the analytical procedure could cause greater differences in the results than were observed between the varieties. Hence, it was considered necessary to investigate first the analytical method.

The soybeans used in this work were Illini variety grown during 1936 near St. Joseph, Illinois. They were cracked, flaked, and then extracted with petroleum ether (boiling range 35 to 60°) in a Soxhlet-type apparatus. The fat-free meal was air dried at room temperature, and ground. In order to avoid the possibility of heat denaturation, the temperature of the meal was kept below 40° C. at all times.

In grinding, it was noted that a burr-type mill would allow the relatively tough, thin hulls to pass through without reducing them to as small a size as desired. A burr grinder is also objectionable because of the danger of altering the proteins by frictional heat. In this work a porcelain ball mill with flint pebbles, or a Wiley mill, was used.

It was observed that the hulls are much harder to grind and that the particles retained on the coarser screens consisted largely of hull fragments. In order to test the solubility of the nitrogen compounds in the hulls, a sample of hulls was carefully separated by hand from oil-free flakes. An analysis of the hulls is presented in Table I and the amount of nitrogenous compounds extracted is shown in Table III. Although the hulls comprise about 6 to 8% of the total weight of the soybean (see Bailey, Capen, and LeClerc, 1935), they contain less than 1% of the total nitrogen, and as shown in Table III this small amount is not easily dispersed in water. These factors coupled with the tendency of the hulls to segregate make it imperative that thorough mixing and careful sampling be employed if concordant results are desired.

Before investigating the effect of each variable the tentative procedure given below was adopted. This procedure is similar in many respects to the method used by Staker and Gortner (1931) in their work on extracting protein from seeds. A weighed amount, usually 2.5 g., of the fat-free meal was shaken with distilled water at room temperature for 30 minutes on a mechanical shaker. The mixture was then centrifuged for 6 minutes at about 3,000 r.p.m., and the liquid was carefully decanted or filtered into a Kjeldahl flask and analyzed for nitrogen.

The residue was again shaken with another portion of water for an equal length of time, and the liquid was analyzed as before. This process was repeated as many times as was considered necessary (usually three).

Following this general procedure, an investigation was made of the effect of the size of the meal particles, the solvent-meal ratio, temperature, time, and separation of the meal residue from the suspensoid.

1. *Size of meal particle.* Five different sizes of meal were prepared from the same batch of flakes. Table I lists the moisture and nitrogen content of each of these five sizes and demonstrates that the grinding and sifting process to which they were subjected did not appreciably change the samples in these respects. The results of duplicate determinations are given in Tables I and III to indicate the precision obtained.

Size No. 1 meal consisted of the whole flakes, including hulls, and was the batch from which the other sizes were obtained by grinding. A screen analysis of the meal prepared in the Wiley mill is given in Table II. Sizes Nos. 2, 3, and 3A were prepared by means of a Wiley

TABLE I

ANALYSIS OF SAMPLES OF SOYBEAN MEAL PREPARED FOR EXTRACTION

Size number and description of particles	Moisture	Nitrogen	Total nitrogen in 2.5 g. sample
	%	%	*Mg.*
Hulls only	10.35	1.44	36.0
No. 1, flakes (including hulls)	9.46	8.19	204.0
	9.48	8.14	
No. 2, Wiley, through 2-mm. openings	9.53	8.05	200.3
	9.51	7.98	
No. 3, Wiley, through 1-mm. openings	9.39	8.21	204.5
	9.41	8.15	
No. 4, ball mill, through 100-mesh screen	9.42	8.21	204.3
(.147-mm. openings)	9.42	8.13	
No. 5, ball mill, through 200-mesh screen	9.38	8.17	203.8
(.074-mm. openings)	9.55	8.14	
No. 6, wet grind in ball mill, No. 4 meal	9.42	8.21	204.3
used	9.42	8.13	

mill using the 2, 1, and .5 mm. screens, respectively. Sizes Nos. 4 and 5 were obtained by grinding in a ball mill until all the material passed through 100- and 200-mesh screens, respectively. During the ball mill grinding the meal was sifted every few hours, the portion that had passed through the specified sieve being removed each time. It required about 24 hours to grind a sample until it would all pass through the 100-mesh screen and 48 hours to reduce to the 200-mesh size.

In addition to these five samples one run was made (No. 6 in Table III) by wet grinding in the ball mill. A 10-g. sample of size No. 4

meal plus 400 ml. of water was ground for 4½ hours. The mixture was then centrifuged, filtered, and a 100 ml. aliquot, representing 2.5 g. of meal, taken. Table III shows the amount of nitrogen extracted from these various-sized meals.

TABLE II

SCREEN ANALYSIS OF 100-GRAM SAMPLES OF SOYBEAN MEAL

Size	Meal retained on mesh size—					Meal passed through mesh size—	Total recovered from sample
	20	35	60	80	100	100	
	G.	*G.*	*G.*	*G.*	*G.*	*G.*	*G.*
No. 2	0.6	34.5	38.7	6.7	2.2	17.1	99.8
No. 3	None	13.4	47.2	14.0	4.7	20.5	99.8
No. 3A	None	None	1.1	3.4	6.3	88.4	99.2

TABLE III

NITROGEN EXTRACTED BY 100 ML. OF WATER FROM 2.50-GRAM SAMPLES OF SOYBEAN MEAL OF VARIOUS-SIZED PARTICLES AS DESCRIBED IN TABLE I

Number of extraction	Nitrogen extracted from samples designated—						
	Hulls	1	2	3	4	5	6
	Mg.	*Mg.*	*Mg.*	*Mg.*	*Mg.*	*Mg.*	*Mg.*
First	8.2	130.0	139.5	146.5	165.3	160.8	189.1
		133.2	139.5	148.7	165.7	161.3	189.2
Second		27.6	23.4	20.9	17.8	17.1	—
		27.6	22.0	22.3	18.1	17.8	—
Third		5.6	4.2	4.2	3.8	2.2	—
		6.3	4.0	4.5	3.7	2.4	—
Total [1]		165.1	166.3	173.5	187.1	180.7	189.1
Total extracted, %		80.9	83.0	84.8	91.6	88.7	92.6

[1] Results with duplicate samples were averaged and then totaled.

From the data given in Table III it may be readily seen that the size of the meal particle has an appreciable effect upon the amount of protein dispersed. As would be expected, the solubility increases with a decrease in particle size except that the 200-mesh meal is slightly less soluble than the 100-mesh. This unexpected result can be accounted for on the basis that the long pounding in the ball mill, necessary to reduce the meal to 200-mesh, has caused a denaturation of the protein. Smith, Circle, and Brother (1938) have noted a similar effect upon the solubility in salt solutions.

On the basis of the greatest amount of nitrogen extracted with a minimum of labor, the method of wet grinding in a ball mill is the most efficient. Although this procedure apparently has not been employed

in analytical studies on the extraction of seed proteins, it appears to offer distinct advantages for this purpose. A very thorough grinding action is obtained and the liquid prevents local overheating and mechanical shock, both of which apparently cause denaturation.

Of the meal particle sizes studied, size No. 4 (100 mesh) was the most satisfactory. It not only yielded a large amount of protein in dispersion but it was also much easier to throw down in centrifuging, thus permitting a clean separation by decantation. The difference between size No. 4 and coarser meal is appreciable in this respect and is of itself adequate reason for adopting the finer size. In the dry grinding of soybean meal it is not practical to attain a limit of fineness more than about 100-mesh. This size meal was used in all subsequent work reported in this paper. Size No. 3A (.5 mm. Wiley) has been found to give practically the same results (see Smith, Circle, and Brother, 1938) as 100-mesh meal. This is to be expected from its screen analysis as shown in Table II.

A few preliminary experiments using 5 and 10% sodium chloride solutions showed that these conclusions hold equally well for salt solutions. It was also noted that pure water removes slightly more nitrogen than the salt solutions tried. Quantitative data on this subject will appear in another publication (Smith, Circle, and Brother, 1938) from this laboratory.

2. *Solvent meal ratio.* In consideration of the analytical procedure involved it was decided that a 2.5-g. sample of meal was best suited for the purpose. A survey of the literature showed wide variations in meal to water or salt solution ratios, with no data to justify the procedure in this respect.

Table IV indicates that there is nothing critical about the meal-water ratio, but some limits may be given. For quantities of water less than 50 ml. the resulting mixture is too viscous to centrifuge or filter readily, and the number of extractions required to remove the

TABLE IV

PERCENT NITROGEN EXTRACTED FROM SOYBEAN MEAL BY VARYING AMOUNTS OF WATER

Number of extraction	Nitrogen extracted by water in amount of—				
	25 ml.	50 ml.	100 ml.	200 ml.	400 ml.
	%	%	%	%	%
First	59.4	73.6	81.0	80.8	82.7
Second	22.4	13.9	9.3	10.1	5.5
Third	5.8	4.0	1.9	1.1	.6
Fourth	1.6	.6	.2	—	—
Total	89.2	92.1	92.4	92.0	88.8

same percentage of protein is greater. On the other hand, large amounts of water did not remove appreciably more nitrogen and are difficult to handle in the nitrogen analysis. It is concluded that a ratio of water to meal of 100 to 2.5 by weight is satisfactory for this extraction. Should it be desirable to keep the total amount of liquid as low as possible, a greater number of extractions with small portions may be used. For example, four 25-ml. portions extracted as much protein as did three 400-ml. portions with a difference of 1,100 ml. in the water used. A comparison of the first extractions, however, shows that the 400-ml. portion removes considerably more than smaller amounts.

In making successive extractions such as described, there will always be some water remaining with the meal. This was from 7 to 10 ml. for 2.5 g. of meal. It is obvious that this water has dispersed its share of protein and that this fraction is carried over to appear in subsequent extractions. In no case investigated does this carry-over account for the total recovered from following extractions although it is responsible for a large share.

In order to determine the effect of moisture content, two samples of No. 4 meal were placed in desiccators. One desiccator contained anhydrous calcium chloride, the other a sulphuric acid water mixture adjusted to maintain a relative humidity of 75%. After three days one sample contained 2.93% and the other 15.61% moisture. The percentage nitrogen extracted from each of these samples was within 2% of that reported in Table III for meal containing 9.42% moisture. Hence, it is concluded that within this range the moisture content of the meal is not a factor influencing the extraction of nitrogenous compounds with water.

3. *Temperature.* Table V shows the effect of temperature variation on the amount of nitrogenous compounds dispersed.

TABLE V

PERCENT NITROGEN EXTRACTED AT DIFFERENT TEMPERATURES

Temperature	Nitrogen
° C.	%
1.5	71.4
14.0	76.3
28.0	80.0
35.0	81.6
45.0	83.1

It can be readily noted that temperature has a definite effect and that extreme variation in temperature will cause discrepancies in the results. In the range of 15 to 35° C. the variation is about 0.25% nitrogen per degree C. The room temperature in this series of experi-

ments averaged 27° C., and it is estimated that variations of not over 1% have been introduced as a result of this factor

4. *Time.* Table VI indicates the effect of time variations in the shaking period on the amount of nitrogen extracted.

TABLE VI

PERCENT NITROGEN EXTRACTED FROM SOYBEAN MEAL, WITH VARIATIONS IN THE TIME OF SHAKING

Number of extraction	Nitrogen extracted when sample was shaken—							
	1 min.	7½ min.	15 min.	30 min.	60 min.	120 min.	180 min.	240 min.
	%	%	%	%	%	%	%	%
First	71.9	75.6	78.0	81.1	80.7	82.3	83.6	83.9
Second	18.1	14.9	—	8.8	—	8.1	8.2	—
Third	2.6	1.9	—	1.8	—	1.4	1.4	—
Total	92.6	92.4	—	91.7	—	91.8	93.2	—

The length of time spent in shaking, like the water-meal ratio, is not at all critical. The results presented indicate that 1 minute's shaking for each of three extractions will disperse as much protein as 3 hours for each extraction. The amount extracted on the first shaking is considerably greater, however, for the longer interval. It should be remembered that the time spent in shaking was not the entire period that the meal was in contact with water. It was found that about 10 minutes were required from the time shaking was stopped until the liquid had been decanted.

A consideration of the mechanics by which a colloidal sol such as this is formed leads to the conclusion that either the protein particle will float away readily from a broken cell or the cell walls must be ruptured by the softening and swelling action of the solvent. The alternative of a migration through unbroken cell walls is rejected as being improbable for a particle having the size of protein molecules. The rapidity with which the dispersion takes place from 100-mesh meal leads to the conclusion that there are but few cell walls which have escaped damage during the grinding process. This conclusion is not in accord with Davidson (1929) who claims that reduction to 200-mesh is necessary to assure that all the cell walls are broken. His criterion, however, was the tensile strength of an adhesive produced after admixture with other substances. It may be noted from Table III that the second and third extractions yield a greater return of soluble nitrogen compounds from the coarser meal sizes, probably because of the time required by the solvent to force open unbroken or only slightly damaged cells.

5. *Separation of meal residue from suspensoid.* After shaking the the meal with water the problem of separating the liquid from the suspended meal must be solved. Even after centrifuging for 10 minutes at 3,000 r.p.m. one can not be assured that minute meal particles are not still suspended. After centrifuging, the liquid has a slightly turbid appearance but no particles are visible to the unaided eye. Filtering through No. I Whatman filter paper did not remove any additional nitrogen as compared with the liquid decanted from the settled meal. Hence, it was considered justifiable to separate this liquid by simple decantation especially in the case of the 100-mesh meal which settles out very readily upon centrifuging.

To indicate the particle size, the suspensoid after centrifuging for 10 minutes as above was filtered twice through No. I Whatman filter paper and then centrifuged again, this time at 48,000 r.p.m. for 30 minutes in a supercentrifuge. A clear, slimy precipitate was thrown out removing 6.86% of the nitrogen from the liquid. The liquid was considerably clearer after this fraction had been removed. A more detailed study of this subject is contemplated and an attempt will then be made to arrive at a particle size distribution.

Summary

A study has been made of five physical factors influencing the amount of nitrogenous material extracted from soybean meal by water. The data presented may be used as a guide in establishing a procedure for the extraction by water of the proteins from soybean meal.

The ratio of water to meal, the temperature, and the time of extraction were found to exert small effects and the magnitude of these effects has been determined.

The size of meal particles is a very important factor. Meal passing through 100-mesh screen was found to be satisfactory for the purpose of extracting nitrogenous compounds. Wet grinding in a ball mill is suggested as an efficient method of dispersing soybean proteins.

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A PRACTICAL AND SIMPLE METHOD OF RECORDING THE FORM AND POROSITY OF BAKED PRODUCTS

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(Received for publication January 16, 1937)

In carrying out practical baking tests it is useful to keep not only a numerical record of the measurements and assigned scores, but also a pictorial record of the form and porosity of the baked product. Heretofore these characteristics have been recorded by preserving and storing the product wholly or in part, by photographing the cut surface, or by spreading an ink over the surface of a slice and stamping an impression on absorbent paper. The impressions obtained by the latter method, however, are lacking in distinctness because of the distortion of the individual pores by the strong pressure exerted during the stamping operation. Moreover, the outlines of the individual pores are not clearly defined, and because of the uneven surface of the slice, many points in the impression are left blank.

If the primary object of these pictorial methods is to obtain a record of the size and form of the individual pores, the above-mentioned photographic method presents many difficulties, because the poor definition of the pores in the resulting photographs does not permit a practical judging of the baked product or an accurate comparison with Mohs' (1924) [1] scale of porosity. It is also possible to ascertain the corresponding numbers on the Mohs scale either by direct comparison with the cut surface or preferably with the stamped impression, but the results have not been found satisfactory.

[1] Mohs, Karl. The size of the pores in baked bread. Cereal Chem. **1**: 149–151 (1924).

An improved method which the writer has introduced into the practice of the Milling Research Station is based upon making a contact print of a thin slice of the baked product directly upon a contrast photographic paper. A slice is cut from the product at least 12 hours

Brno II

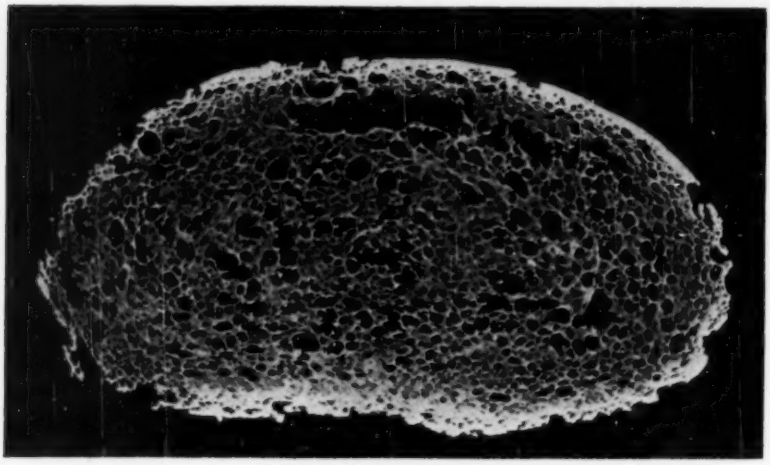

Figure 1

Dregerova OD

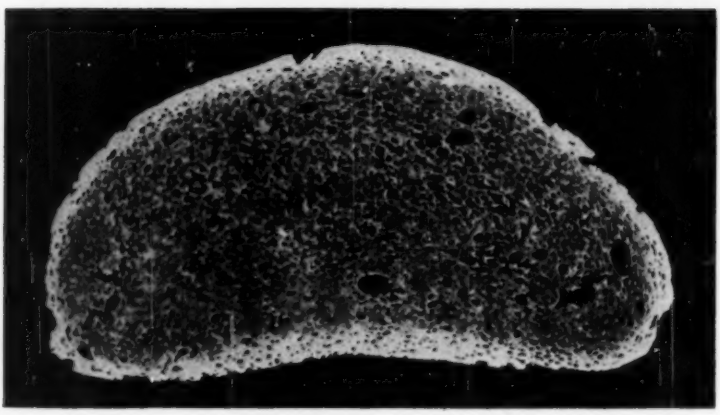

Figure 2

after baking, the smooth cut being made by means of a sharp knife or, preferably, a hand-driven slicing machine, of the type used in restaurants for slicing bread, bacon, etc. The machine is equipped with a cutting disc driven by a gear and a hand wheel. The mounting table can be moved in a vertical direction, and the thickness of the

slice can be regulated at will. The slices are cut to a thickness of 0.50 to 0.75 mm., and the crumb structure should be well preserved. Several slices are taken in this manner and the thinnest one showing the best preserved crumb structure is selected. As the slice is prone to

Perbete

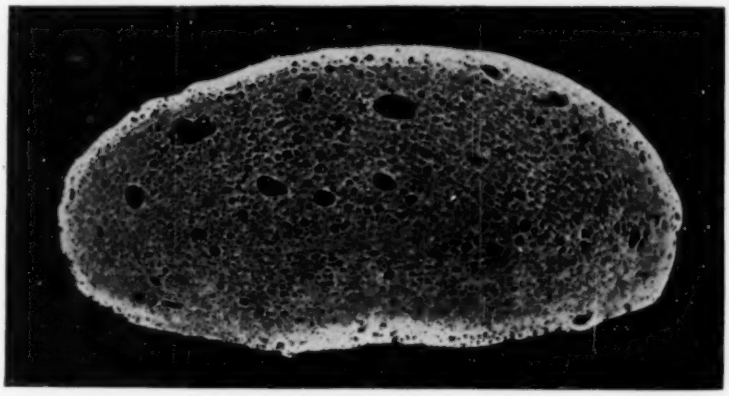

Figure 3

Stocken Slezsko

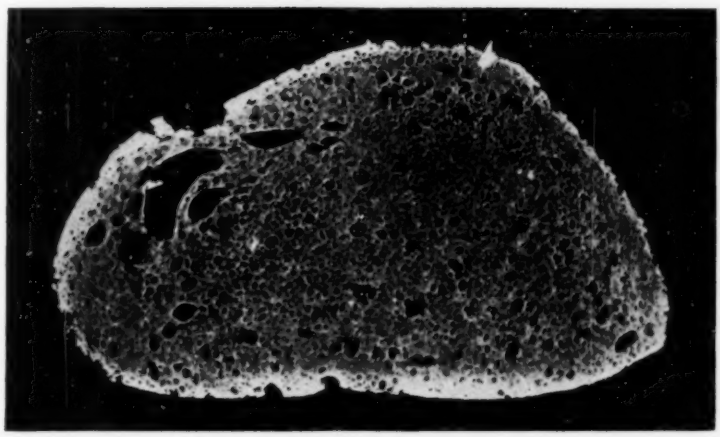

Figure 4

drying and curling, it is immediately placed between two glass plates, the lower of which is covered with a filter paper soaked in a 75% solution of glycerine in water. An excess of the solution must be avoided to prevent the filling of the minute pores of the slice. This method of treatment insures an absolutely flat slice.

The slice soaked in diluted glycerine is now taken to a dark room and transferred to a photographic paper of the proper size, which is placed upon a perfectly flat surface, such as a glass plate. Each slice may be provided with a celluloid label bearing the name and reference number of the sample in India ink. The photographic paper bearing the slice is now exposed to a suitable source of illumination, preferably a frosted lamp, placed 25 to 30 cm. from the paper, and given the proper exposure. The paper is then removed and developed and fixed in the usual manner. The photographic paper best suited for this purpose is a contrast developing paper with glossy or semi-glossy surface.

The procedure as described is very simple, rapid and inexpensive. The prints furnished by this method present a great advantage over those obtained by stamping the impression of the inked slice, as the pores of the baked product are clearly defined and free from any distortion due to strong pressure. The prints (see Figures 1 to 4) show the shape of the loaf as well as the thickness of the crust.

By this method for rapid recording of the structure of baked products in laboratory practice, a number of prints may be collected and used as a basis of comparison in commercial and forensic analysis. A scale of porosity resembling that of Mohs could be set up, using numbers from 1 to 10 to denote increasing pore size. Such an improved scale would permit more accurate and easier comparisons, and the difficulties encountered in attempting to relate imperfect impressions to the individual types of porosity shown in the Mohs scale would be eliminated.

SOME PHOTOMICROGRAPHIC STUDIES OF DOUGH AND BREAD STRUCTURE

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(Received for publication August 7, 1937)

Earlier work on the subject was carried out by Scheffer and published in 1916. Scheffer stated he was unable to discover previous work in this field. He observed that the paraffin and gelatine embedding methods did not give satisfactory results for the cutting of thin sections of bread, and recommended cutting the semi-dried breadstuff by hand microtome using an oblique cutting action. If the sample had hardened or dried out too far, it was softened slightly by keeping in a moist atmosphere containing formaldehyde to prevent mould growth. The section was examined in a mixture of glycerine and water and much information was gained without staining the specimen. The use of polarized light was also recommended. A mixture of methyl green and cresol violet was suggested as particularly suited to the examination of wholemeal bread because the different parts of the bran of the grain are differentially stained by these reagents. Scheffer illustrated his paper with photomicrographs.

The subject was taken further by J. R. Katz and E. Van Teutem who published the results of their investigations in 1917. Bread slices were cut after embedding in gelatine and no photomicrographs were given, the paper being illustrated by diagrams by E. Van Teutem. A gluten structure with starch and yeast embedded was observed. The starch cells adjacent to the air cell surfaces were elongated or "bean" shaped, while those further within the breadstuff appeared to retain their original shape. A comparison with the structure of a cut-steel surface was made, where all the particles appear to be oriented by the flow of the metal before setting. It was further stated that in the bread from correctly fermented doughs, the starch cells present a regular formation, the larger cells being interspersed with smaller cells, while in bread from overripe doughs the degree of dispersion is greater, the starches are more widely separated and all formation is lost. In underripe dough the cells are closer, overlapping, and again show no regular formation. In underfermented and overfermented loaves it is stated that a lower proportion of gluten is to be seen.

The work of Verschoffelt referred to by Katz (1917) showed the development of air layers around the starch cells in old bread and this phenomenon was illustrated diagrammatically by E. Van Teutem. The irregular shape of starch cells in old bread was compared with the rounded cell forms existing in fresh bread.

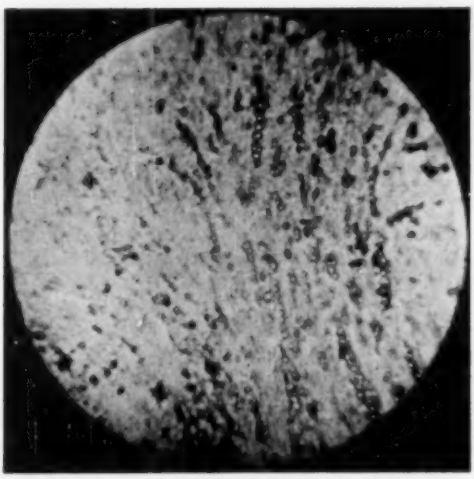

Figure 1. Gluten film (low power). × 100.

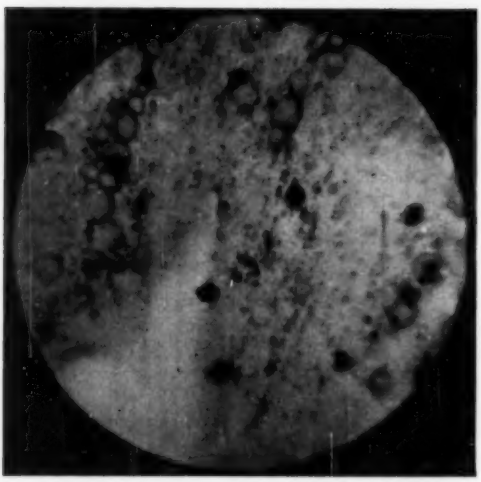

Figure 2. Gluten film (high power). × 400.

The present work was undertaken with the object of observing dough and bread structure, developing different micro-methods of investigation and obtaining photomicrographic records of the observations made. Work on gluten films was first carried out, and Figures 1 and 2 show the gluten films obtained by continued stretching of

hydrated gluten. In Figure 1 (low power), the starch cells were stained with iodine and the film allowed to dry (no cover slip was used). The uniform order of the starch cells in Figure 1 shows the strain to which

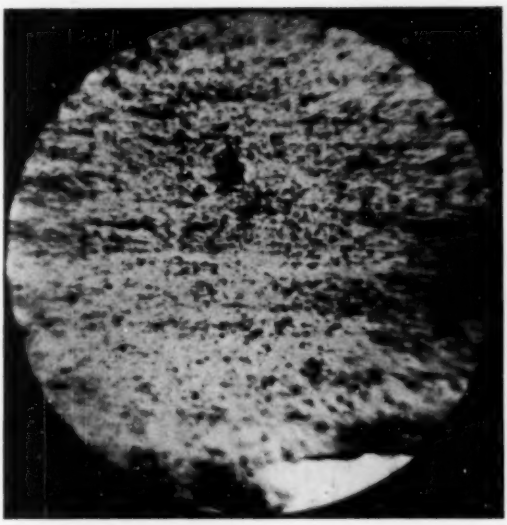

Figure 3. Gluten film (low power). ×60.

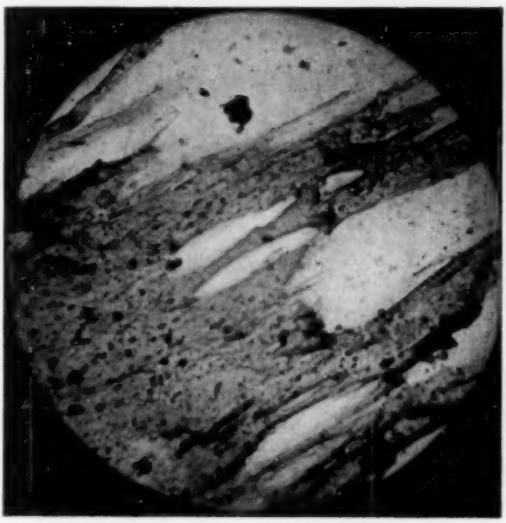

Figure 4. Gluten film (low power). × 60.

the gluten was subjected during stretching. Figure 2 (under higher power) shows the disposition of the starch cells more clearly. It is interesting to note that in Figures 1 and 2 there is little evidence of

the presence of the gluten film. Only in Figure 2, where an edge of the film is shown, is the gluten visible. Figures 1 and 2 are not stained

Figure 5. Sponge strand (low power). × 100.

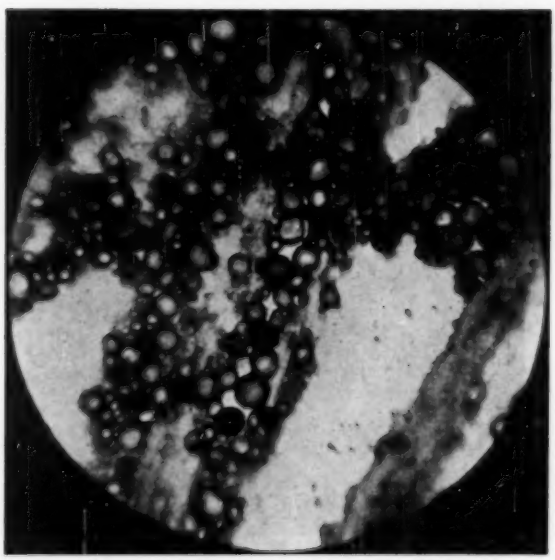

Figure 6. Sponge strand (high power). × 400.

with eosin but even when such a stain is used the presence of a thin gluten film is not easily detected. Figure 3 exhibits the same phenomenon and Figures 1, 2, 3 and 4 support the view that gluten is

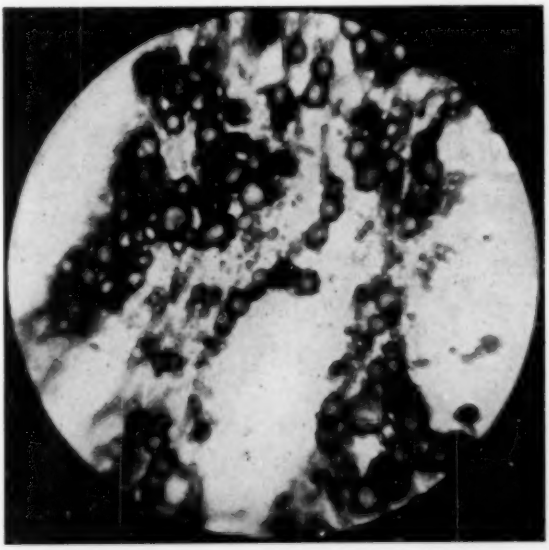

Figure 7a

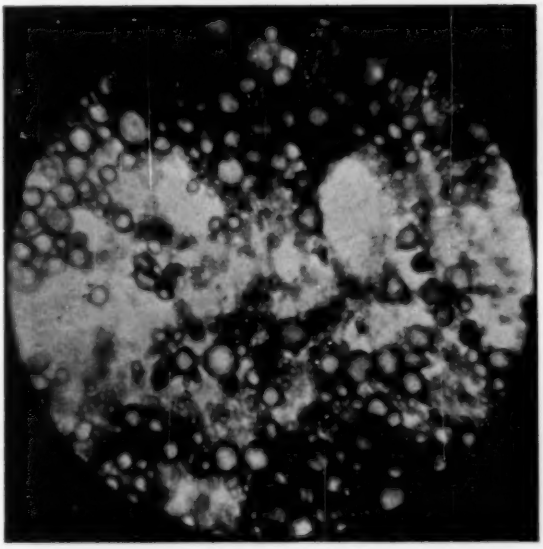

Figure 7

Figure 7. Correctly fermented dough film (high power). × 400.

devoid of organized structure so far as can be ascertained microscopically. Figures 3 and 4 are further low-power photomicrographs

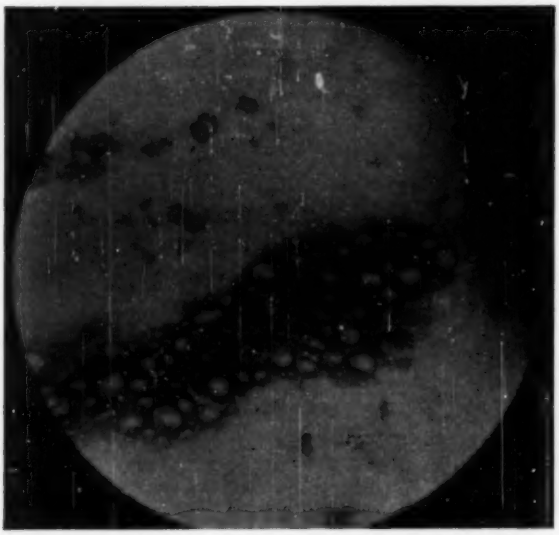

Figure 8. Overfermented dough film (high power). × 400.

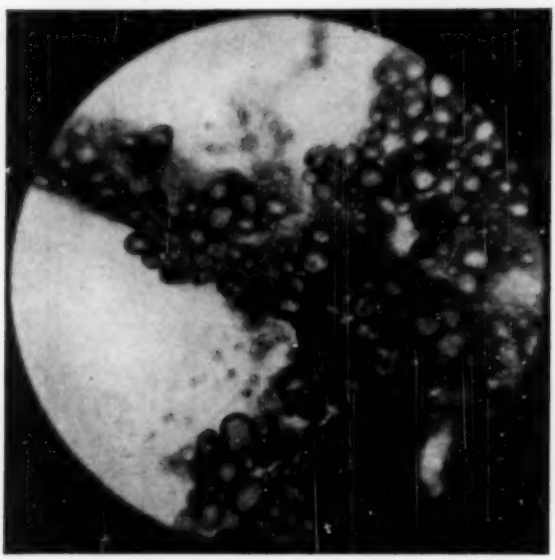

Figure 9. Underfermented dough film (high power). × 400.

showing the pull to which the gluten films were subjected, Figure 4 showing the broken edge of such a film.

The stretching method was applied to sponges and also to doughs.

A sponge was taken just before the dropping point and a thin strand selected and placed on a microslip which gave the results shown in Figures 5 and 6. This was stained with eosin and iodine but received no further treatment; the gluten structure is clearly seen supporting the clustered starch cells, some of which are partially embedded in the gluten mesh along with yeast cells. It will be observed also that some of the starch cells have taken the iodine stain and show up black while the unruptured cells are unstained. Figure 5 shows the low-power field and Figure 6 a section of the same field under higher power.

Figures 7, 7*A*, 8, 9 and 10 were taken from dough films. Figures 7 and 7*A* (high power) show correctly fermented dough. A section was made when the dough was ready for the divider, a short time allow-

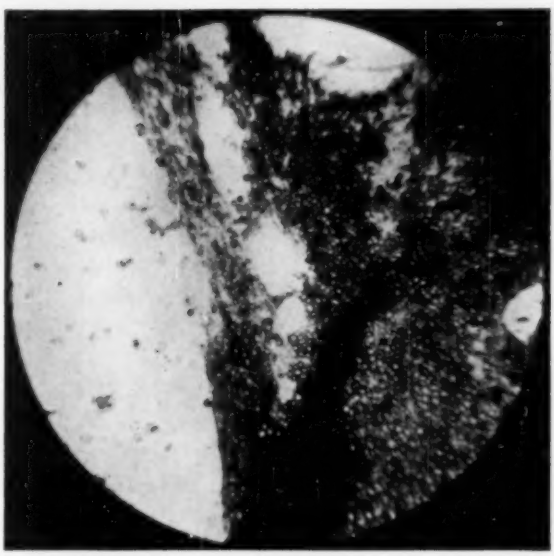

Figure 10. Underfermented dough film (low power). × 100.

ance (15 minutes) being made for the preparation and staining so that the dough was correctly fermented when the photograph was taken. Figure 8 shows the structure of an overfermented dough and Figures 9 and 10 that of an underfermented dough under high and low power, respectively.

The photomicrographs of dough are typical of many fields examined. No evidence was discovered in support of the view that varying periods of fermentation give rise to characteristic changes in the arrangement of starch cells. Regular and irregular starch formations were observed in underfermented, correctly fermented and overfermented doughs and such formations would therefore appear to be of

little value in determining the degree of dough fermentation (see Figures 7, 7*A*, 8, 9 and 10).

It would appear that under the normal conditions existing in a fermenting sponge or dough, a much smaller proportion of the starch is truly "embedded" in the gluten than had been previously considered to be the case.

It is probable that immediately after mixing or processing (dividing moulding, etc.) much starch is embedded in gluten, but as fermentation proceeds the mechanical effect of the gas produced upon the gluten structure brings about a "squeezing out" of the starch cells from the gluten so that the cells adhere to the surface of the gluten mesh in

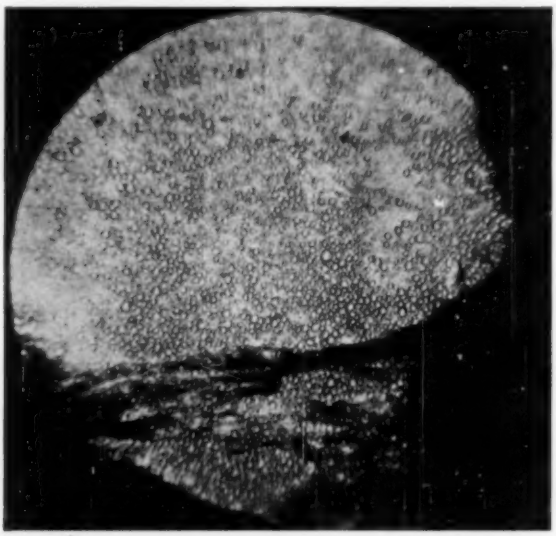

Figure 11. Disturbed dough film (low power). × 100.

increasing proportions, the quantity embedded in the gluten decreasing accordingly.

During the course of the work on sponges and doughs it was found that ordinary methods of staining and preparation were unsuitable because such methods interfered greatly with the true structure of the material examined. Further it was observed that when a sponge or dough film was teazed out with a mounting needle in the presence of a liquid (stain, water or glycerine), starch cells in considerable numbers were readily detached from the gluten structure and dispersed in the liquid. Such cells then concentrated against the first gluten strand, forming a barrier to the flow of the liquid. If such a slide was allowed to dry various starch formations, regular and irregular,

could be obtained and for some time such formations were difficult to account for.

Figure 11 (low power) shows starch cells floated off from their normal position by stain concentrated against a gluten strand. The

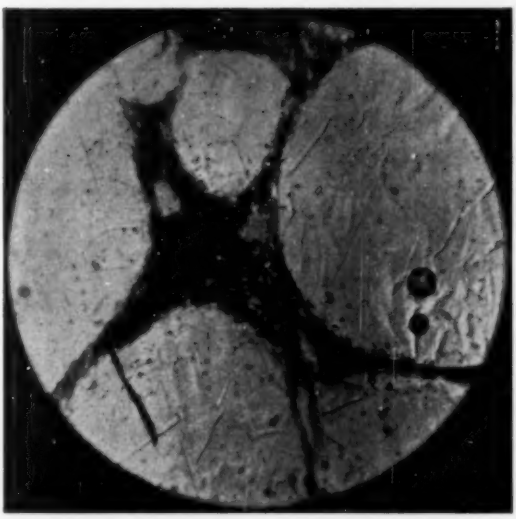

Figure 12. Bread section (low power). × 60.

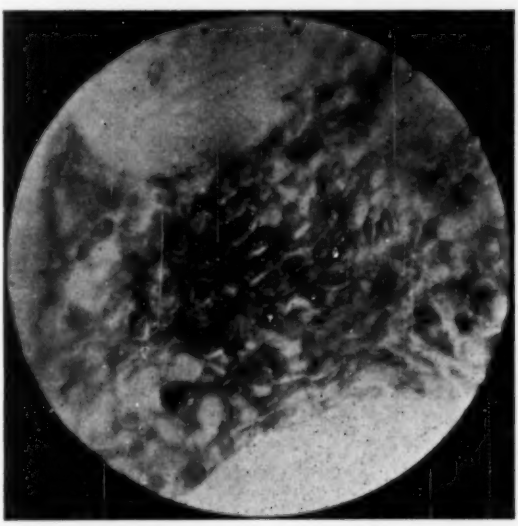

Figure 13. Bread section (high power). × 240.

photograph was taken while the stain was still liquid. Although much of the starch was lost in the manner described many starch cells can be seen adhering to the gluten strand in their original positions, the

proportion dispersed depending upon the amount of manipulation or "teazing out" received by the dough.

It was found to be essential in the work described to observe the material with the minimum of preparation and staining, otherwise distorted results were obtained; the simplest technique produced the most accurate results.

After the experience gained from gluten, sponge and dough films, work on bread was undertaken. Figures 12, 13 and 14 show sections of bread cut by means of the freezing microtome after the small bread cubes had been hardened in formalin, and embedded in increasing gelatine concentrations. Figure 12 (low power) shows the starch cells stained with iodine within the texture of the bread, while Figures 13 and 14 show sections of the field under higher power. The gluten

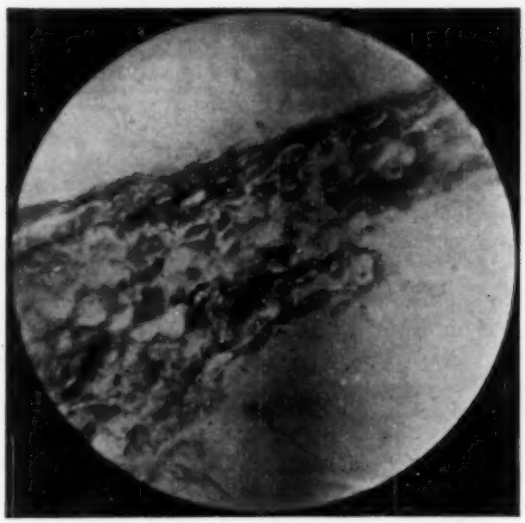

Figure 14. Bread section (high power). × 240.

structure is shown up by the eosin stain, and the somewhat deformed shapes of the starch cells and the "bean shape" referred to by Katz are also visible.

It was found difficult to cut sections sufficiently thin for photomicrographic purposes and it was also thought that the gelatine technique could not fail to give distorted results due to the prolonged soaking period and the formalin dehydration. Teazing out the breadstuff was therefore tried because under high power the breadstuff structure is observed and not the bread texture. It was thought that the manipulation of the breadstuff with the mounting needle might destroy the general texture but that the micro-structure of the breadstuff would remain undisturbed. The teazing out or disintegration of

the bread was only carried out to a degree which made microscopic observation under high power possible. Figure 15 shows the result of such teazing. Swollen starch cells and gluten masses are present but there is no sign of organized structure and the technique was abandoned because further work showed that even slight manipulation caused a disproportionate degree of disturbance, the phenomena observed being very similar to those previously referred to and illustrated in Figure 11. If a small amount of water or glycerine was used the disturbing effect of manipulation was intensified. A comparison of Figure 15 with the photomicrographs of sponge and dough structure

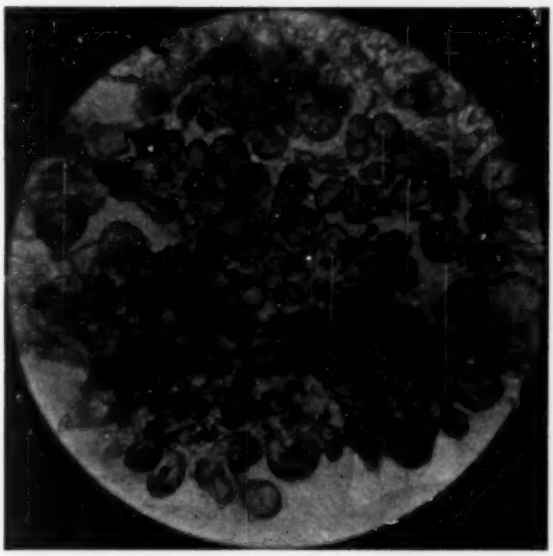

Figure 15. Bread section after "teazing." × 400.

(Figures 5, 6, etc.) illustrates the increase in size of the starch cells due to baking, the high-power magnifications being identical.

Sections of one-day-old bread were then cut with a hand microtome, the bread slice being held in pith for support, these sections were stained with eosin and iodine and were examined without further treatment. The result obtained is shown in Figure 16. Only a small section of the field is in focus but that section shows the gluten strand formation in the bread and also the presence of overlapping starch cells. The resemblance of this small section to the stretched gluten film shown in Figure 3 may be noted.

Figure 17 shows another hand-cut section stained with eosin and iodine in dilute aqueous solution. The swollen and ruptured cells with the dark-stained gluten may be observed. The difference between Figures 12, 13 and 14 and Figure 17 is marked. The latter

photomicrograph shows no sign of elongated starch cells; it may be that this peculiar "bean" shape is due to one or another of the processes employed in the gelatine embedding.

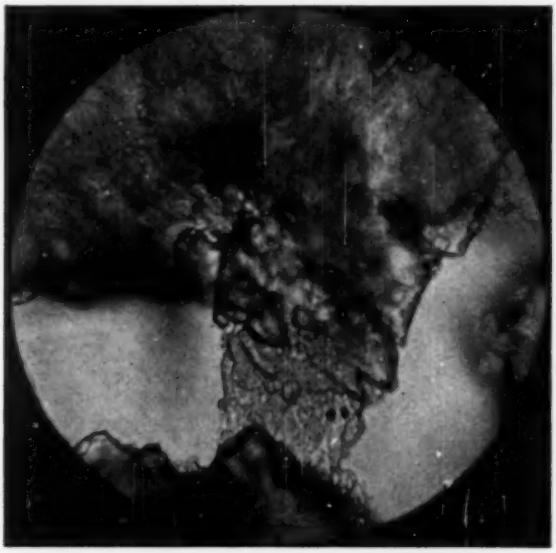

Figure 16. Bread section (low power). × 100.

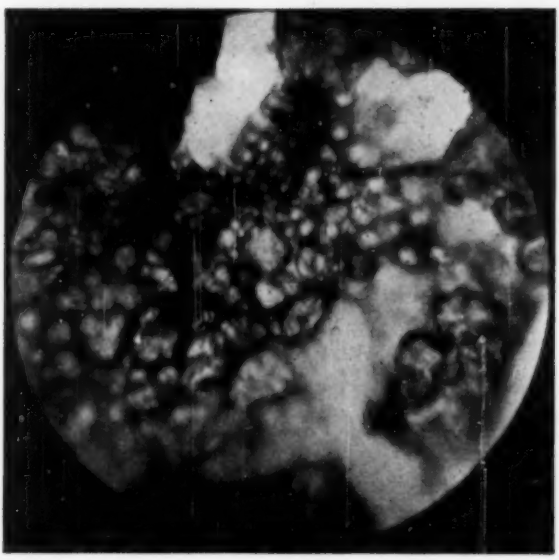

Figure 17. Bread section (high power). × 400.

A number of experiments were carried out using various methods of preparing thin microscopic sections of bread. Figure 18 shows a

Figure 18. Bread section. Untreated. (Low power). × 100.

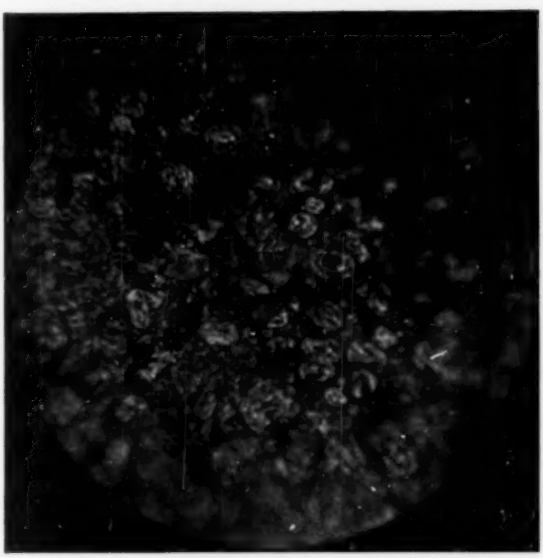

Figure 19. Bread section (high power). × 400.

section of bread cut with a sharp knife. The thin slice of bread was cut from the loaf when approximately 12 hours old, the thinnest portion of this slice was selected, placed on a microslip and photo-

graphed without receiving any further treatment. The bread texture or vesiculation of the crumb is clearly seen.

Figure 19 shows bread structure as obtained by carefully selecting a section direct from the loaf and staining with iodine and eosin. The ruptured and unruptured starch can be seen; also overlapping starch cells and small yeast cells are visible.

Photomicrographic Procedure

A quarter plate camera was erected vertically above the microscope. Illumination was provided by a 60-watt gas-filled lamp 15 cm. distant from the microscope mirror. A substage condenser and mechanical stage were fitted. The plates used had a speed of 700 H and D, exposures ranging from 1 second to 6 seconds. The best results were obtained with 3 seconds exposure on the lower powers, and 5 seconds on the higher powers. For the low-power work 1 inch and ⅔ inch objectives with either No. 2 or No. 4 oculars were used while a ⅙ inch objective with similar oculars was found most suitable for high-power work.

Summary

Earlier work by Scheffer and Katz is reviewed.

Work on gluten films is described.

A study of sponge and dough films suggests that, as fermentation proceeds, the mechanical effect upon the gluten of the gas produced squeezes out the starch cells so that they adhere to the gluten surfaces in increasing quantities.

The material is best observed with the minimum of preparation, otherwise distorted results may be obtained.

No apparent differences are observed in the structure of dough at varying stages of fermentation.

Experimental work on the preparation of dough and bread sections is discussed.

Acknowledgment

The writers are greatly indebted to F. K. Birkett for the preparation of the gelatine-embedded bread sections.

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A STUDY OF VARIOUS CHARACTERISTICS OF MILL-STREAM FLOURS AND THEIR RELATION TO LOAF VOLUME

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(Received for publication January 24, 1938)

Introduction

The importance of the various quality characteristics of mill stream flours needs no emphasizing to the mill chemist. These flours are the materials, as it were, from which the finished flour is constructed, and, as the quality of an edifice is very largely determined by the characteristics of its material, so the baking value of a flour is conditioned by the attributes of its constituent mill streams. These mill streams originate primarily from different portions of the wheat kernel: the flours of better color and low ash come from the more central portion of the endosperm, but with the gradual inclusion of material from the outer or branny layer, the color decreases and the ash content rises markedly. Even the best flours, of course, contain a small proportion of lower quality particles, such as bran, but these are present in relatively minute quantities. As the tail of the milling system is approached these impurities increase rapidly with a consequent influence upon flour quality. The situation in this instance is quite different from that encountered when dealing with flours milled from individual samples of wheat. The latter case is the one which cereal chemists are usually called upon to deal with, and quality variations may here be attributed to genetical or environmental differences in the wheat.

Bailey (1925) listed several analyses of roller mill streams published by Richardson (1884), Jacobs (approximately 1915), Swanson, Willard, and Fitz (1915), and Weaver (1921). The low ash content of the purified middlings was noted by these workers, who remarked also upon the tendency of this constituent to increase as the end of the milling system was approached. The protein percentages tended to increase in progressing through the milling system, while the percentage of dry crude gluten paralleled the nitrogen content rather closely. Loaves of high quality were baked from the break flours despite the lower gluten quality assigned these flours. Swanson and his coworkers found the highest color and texture in the first to sixth

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middlings flours respectively, and Weaver obtained the best bread from the streams lowest in ash. Gluten content was found to increase with ash in the middlings flours. This relationship did not exist in the break flours.

Pascoe, Gortner, and Sherwood (1930) examined a series of mill-stream flours from the Minnesota State Mill and found a negligible correlation to exist between flour protein and loaf volume as obtained by the Standard Basic Procedure advocated by Blish (1928). A relatively high correlation was obtained between these variables in a series of samples milled from different lots of wheat of the same grade. Marked variability was evident in the saccharogenic activities of the various mill-stream flours. The germ content was especially indicative of this activity, as pointed out by these researchers. The third break was lowest and the sizing flours highest in this respect. There was little relationship noted between the peptizable protein fractions and loaf volume.

Harris (1930) published the results of a study made on mill-stream flours produced from hard red spring wheat. This investigation dealt principally with the relations between flour protein, peptization, and baking strength. While total flour protein and loaf volume were highly related when the 12 best flours were considered, this relationship decreased to a point of little practical importance when the lower quality streams were included in the study. The use of a flour "improver" (3% malt and 0.5% Arkady) did not appear to be justified by the results obtained when compared with corresponding values yielded by the standard basic formula. A consideration of quantity of protein peptized in addition to a knowledge of total protein yielded slightly more information with respect to loaf volume for the entire set of twenty flours.

Rich (1934) thought that the presence of germ particles induced low grade quality in Canadian hard red spring wheat mill-stream flours, and attributed the beneficial effects of bleaching to oxidation of some germ constituent.

Later, Harris (1937, 1938) indicated the probable importance of the initial fraction precipitated from gluten dispersions in 10% sodium salicylate solutions by concentrated $MgSO_4$ solution as an index of the baking quality of North Dakota durum and hard red spring wheat flours. A separation of flours produced on an experimental mill to conform approximately to commercial mill streams was also studied, and the results obtained appeared to show a similar relationship in this instance also. The evidence obtained in the latter instance, due to the small number of samples involved as well as to the method of separating the various flours, was not considered sufficiently conclusive

by the author. Accordingly an investigation was undertaken to ascertain more definitely the relationships existing between the different factors of baking quality and loaf volume in a series of mill-stream flours produced in a modern commercial flour mill from a blend of sound bread wheat.

Experimental

Material and Methods

The mill streams described originated from various portions of the wheat kernels comprising a wheat mix of 1937-crop hard red spring wheat, the entire set or various combinations going to make up a finished product. This mix was cleaned, washed, and tempered in the normal procedure used in most mills and gave results as shown in the various reductions.

Two samples were taken direct from each stream; one in a small, air-tight container to secure analysis as actually milled; the other a two-pound bag sample for baking and further chemical examination. The total sampling time was approximately 30 minutes, which insured that no significant change in milling conditions occurred during the sampling period.

Moisture determinations on these samples were made by the air-oven method described on page 29 of the A. A. C. C. Cereal Laboratory Methods (1935).

Ash determinations were made by a modified direct burning method. The usual 5 g. of flour were used and a standard electric muffle furnace was used for burning. To shorten the burning time, a small amount of oxygen was kept flowing into the ignition chamber of the furnace through a very fine jet (approximately the size of a small hypodermic needle). To insure a constant flow of oxygen, about 10 pounds of pressure per square inch were maintained on the oxygen line. The jet is constructed in such a way that it disperses the oxygen into the oven without blowing any flour from the dishes. Flat silica dishes, of 15 c.c. capacity, 47 mm. diameter, and 13 mm. depth, were used for ashing the flour. The flour was ignited at 800° F. and the volatile matter burnt off before closing the oven. The oven was then closed and raised to 1100° F., and burning continued for two hours. This method has been found to approximate closely results obtained by the usual 5 to 6 hour method.

Protein determinations ($N \times 5.7$) were made on the samples according to the standard method outlined in Cereal Laboratory Methods.

The diastatic activity of the flours was determined using the Blish-Sandstedt (1933) method. The results are reported as milli-

grams of maltose produced in 1 hour from 10 g. of flour at 30° C. The resistivity of the starch substrate was also found, using a modification of the procedure developed by Malloch (1929), and contained in Cereal Laboratory Methods, pages 58–59. This modification was used by Shellenberger and Bailey (1936) in an investigation of the malting and biochemical properties of different varieties of barley, and differs chiefly from the original Malloch method in that three washings are made with distilled water without buffer solution, and in the digestion, which is carried out at the same temperature as the determination of diastatic activity (30° C.) rather than 27° C. Five grams of flour are used instead of 10 g. as proposed by Malloch. The glutens were washed from these flours by the procedure followed by the senior author in previous fractionation work (Harris, *loc. cit.*), and dispersed in 10% sodium salicylate solution. The gluten protein thrown down by one addition of concentrated $MgSO_4$ solution was determined using the technique and concentrations developed in this previous work.

The flours were baked by two procedures. One was essentially the Standard Basic Procedure mentioned by Blish (1928) as a reference method. The method used in this instance, however, contained 5% sucrose instead of 3%, as called for in the original basic procedure. The second method employed was the malt-phosphate-bromate formula in which 0.3% Lintner malt, 0.1% ammonium dihydrogen phosphate and 0.001% potassium bromate were included in addition to the ingredients used in the basic method. Three-hour fermentation periods were used throughout, and a two-minute mix in the Hobart-Swanson. The second method was used to bring out the full potentialities of the flours, and it was thought it would be especially valuable when baking the high gluten break flours. Harris noted a decided increase in the loaf volume of break flours when baking a series of hard red spring wheat mill-stream flours with an "improver" formula consisting of 3% malt and 0.5% Arkady. A marked increase in volume with the malt-phosphate-bromate formula was also remarked by Harris and Sanderson (1938) when working with hard red spring wheats grown in North Dakota in 1937.

Discussion

In Table I are shown the flour description, moisture, ash, protein, gluten content, and diastatic activity of the 27 samples included in this study. The moisture is highest in the initial break flours, tending to decrease as the tail of the milling system is approached. The duster flours contain material adhering to the bran which is relatively high in moisture and consequently raises the moisture content of these

flours above the average level for flours produced near the tail of the mill. The first four middlings flours are the lowest in ash, while the fifth break, low grade, and duster flours are very high in this constituent. A rather regular increase in ash is evident in progressing through the reduction system. This increase in ash is roughly paralleled by an increase in the protein and gluten content, which can doubtless be explained by the contamination of the lower grade of flours with material adjacent to the bran layer, which contains a

TABLE I

COMPOSITION OF FLOUR MILL STREAMS USED IN THE INVESTIGATION

(13.5% moisture basis)

Serial number	Mill stream	Moisture	Ash	Protein (N ×5.7)	Wet crude gluten	Protein (N ×5.7) in gluten	Gluten protein in flour	Diastatic activity
		%	%	%	%	%	%	*Mg.*
37–11– 4	1st break	16.2	0.50	13.3	47.8	26.1	12.4	188.9
5	2nd break	16.1	.44	14.8	51.5	27.0	13.9	137.3
6	3rd break	15.3	.55	16.6	63.0	25.5	16.1	135.8
7	4th break	15.2	.75	17.8	68.2	24.8	16.9	145.9
8	5th break	14.4	1.16	18.9	65.2	25.3	16.5	220.9
9	3rd break, No. 1	13.9	.43	12.3	43.7	25.4	11.1	214.4
10	3rd break, No. 2	14.2	.39	13.2	47.7	25.4	12.1	198.2
11	1st sizing	14.8	.63	12.1	41.3	25.5	10.5	340.1
12	2nd sizing	14.7	.40	11.8	41.5	27.2	11.3	418.4
13	3rd sizing	13.0	.47	11.8	40.5	26.3	10.6	237.4
14	1st middlings	14.3	.35	12.0	40.3	26.7	10.8	247.1
15	2nd middlings	14.3	.34	11.9	40.4	26.8	10.8	288.1
16	3rd middlings	13.6	.35	12.3	40.6	26.4	10.7	255.1
17	4th middlings	12.6	.38	12.8	42.6	26.9	11.4	257.7
18	5th middlings	12.8	.49	13.4	46.8	25.4	11.9	256.6
19	6th middlings	13.7	.54	14.0	49.7	25.4	12.6	220.7
20	7th middlings	13.3	.60	14.3	48.6	25.1	12.2	216.9
21	8th middlings	12.5	.67	14.1	48.3	25.9	12.5	244.8
22	1st tailings	13.7	.78	13.8	46.9	26.0	12.2	269.2
23	2nd tailings	13.6	1.08	15.5	54.2	24.9	13.5	235.8
24	3rd tailings	13.1	.74	12.9	48.3	26.2	12.6	261.6
25	1st low grade	12.4	.93	14.6	47.9	25.6	12.3	264.8
26	2nd low grade	11.6	.98	15.1	49.7	25.1	12.5	282.5
27	3rd low grade	12.1	1.15	15.5	49.0	25.1	12.3	280.5
28	Dust reel	13.3	.57	12.1	38.6	26.7	10.3	390.7
29	B & S duster top	14.3	1.47	16.8	56.2	25.3	14.2	329.7
30	B & S duster bottom	14.1	1.49	17.0	59.1	25.6	15.1	267.4

higher proportion of mineral and nitrogenous constituents. The saccharogenic activity is high in the sizing flours as noted by Pascoe, Gortner and Sherwood (1930) and was explained by them as due to the presence of germ material in these flours. The dust reel flour is also extremely high in diastatic activity and low in ash.

In Table II the fractionation and baking data are presented, with the results of the determination of the starch substrate resistivity to

enzymic attack. There does not appear to be any great variability in the solubility of the gluten proteins in 10% sodium salicylate among the different flours, the proteins of the lower quality stream apparently having no greater solubility in this medium than the best flours. Similarly, the quantity of protein fractionated by $MgSO_4$ does not differ markedly from sample to sample in this series. This fraction, when computed as percent of flour, appears to vary significantly with flour strength, but this is caused by differences in the protein or gluten content of the flours, as the gluten fraction in the flour is largely influenced by the percent of crude protein present. Large differences in the starch resistivity are evident. These values are extremely low for the first two sizing flours, which were high in diastatic activity. It would appear accordingly that the high saccharogenic activity of these flours is due largely to a lower starch resistance to hydrolysis rather than to an increase in enzymic activity. The same reasoning applies to the duster flour. It is probable that the starch granules in this sample had been injured by severe grinding and mechanical treatment in the mill with consequent impairment of resistance to hydrolysis by the diastatic enzymes. The Standard Basic Method did not differentiate as sharply among the flours in respect to loaf volume as the malt-phosphate-bromate baking. The latter method brought out in a striking manner the potential baking quality of the first five break flours. This reaction corresponds essentially to the results obtained by other workers and already cited in this paper. The break flours produced loaves of darker color and lower texture rating than the better middlings flours. Loaf volume data only were included in Table II, as this characteristic of the baked loaf is subject to quantitative measurement.

The statistical constants computed from the data are shown in Table III. The range in diastatic activity and starch resistivity is extremely large, reflecting significant differences in these attributes from sample to sample. Flour ash is also extremely variable, as would be expected in material of this nature. Large variations are evident in flour protein, crude wet gluten, and malt-phosphate-bromate loaf volume. The quantity of protein fractionated, and the Standard Basic loaf volume, on the other hand, have relatively small variability.

The correlation coefficients calculated from results of the various determinations are presented in Table IV, with tests of significance. Relatively high positive correlations are shown between loaf volume and flour protein, loaf volume and wet crude gluten, and loaf volume and gluten protein in the flour. No significant correlation was demonstrated between loaf volume and the protein fraction. This

TABLE II

PROTEIN FRACTIONATION DATA, STARCH RESISTIVITY, AND LOAF VOLUME OF THE MILL-STREAM FLOURS

Serial number	Soluble protein mg. per 100 c.c.	Protein removed by $MgSO_4$ per 100 c.c.	Protein removed by $MgSO_4$ as percent of flour	Starch resistivity	Loaf volume (13.5% moisture basis)	
					Standard basic	Malt-phosphate-bromate
		Mg.	%		*C.c.*	*C.c.*
37–11– 4	490	134.0	3.20	751	568	790
5	481	139.6	3.59	685	566	840
6	475	137.4	4.33	605	584	960
7	478	148.2	5.05	599	581	920
8	496	151.0	4.92	579	566	825
9	479	148.8	3.25	445	538	634
10	479	139.0	3.31	442	552	706
11	450	126.0	2.60	356	560	650
12	473	141.4	2.93	249	544	560
13	476	112.2	2.27	515	526	565
14	481	148.2	2.99	324	522	555
15	473	133.8	2.70	416	520	550
16	496	152.6	3.10	324	518	605
17	504	156.0	3.32	385	536	605
18	485	147.0	3.44	360	555	710
19	484	158.4	3.94	509	548	725
20	496	157.2	3.82	342	562	739
21	499	155.6	3.76	463	548	704
22	484	149.8	3.51	377	534	718
23	464	140.2	3.80	533	555	715
24	467	139.0	3.36	390	552	692
25	499	156.6	3.75	380	532	660
26	487	139.0	3.45	306	532	674
27	487	140.8	3.45	435	553	702
28	473	134.4	2.59	235	532	535
29	493	159.0	4.47	477	595	750
30	493	149.4	4.41	501	585	746

TABLE III

STATISTICAL CONSTANTS CALCULATED FOR THE VARIOUS DETERMINATIONS

Determination	Mean	Minimum	Maximum	Standard deviation	Coefficient of variability
Crude flour protein, %	14.1	11.8	18.9	1.96	13.90
Wet crude gluten, %	48.8	38.6	68.2	7.73	15.84
Gluten protein in flour, %	12.6	10.3	16.9	1.80	14.29
Protein removed by $MgSO_4$, mg.	144.2	112.2	159.0	11.29	7.83
Diastatic activity, mg. maltose	252.1	135.8	418.4	65.14	25.84
Starch resistivity	443.8	235	751	123.68	27.87
Flour ash, %	0.69	0.34	1.49	.330	47.80
Protein solubility in sodium salicylate, mg.	483.0	450	504	94.7	19.61
Loaf volume (Standard Basic), c.c.	550.5	518	584	20.55	3.73
Loaf volume (malt-phosphate-bromate), c.c.	697.59	550	960	105.84	15.17

fraction appears to be relatively constant in the gluten of flours originating in various parts of the wheat kernel. A negative relationship is apparent between malt-phosphate-bromate loaf volume and diastatic activity. This is evidently not due to the higher saccharogenic activity of low grade flours, as no significant correlation was found between diastatic activity and flour ash. A positive relationship between loaf volume and starch resistivity was noted. Protein solubility had no connection with loaf volume. A high positive correlation existed between flour protein and wet crude gluten, as would be expected, while high protein millstream flours tend to be also high in ash. No relation was evident between flour protein and gluten protein solubility or between flour protein and the quantity of gluten protein fractionated by $MgSO_4$.

TABLE IV

CORRELATION COEFFICIENTS COMPUTED FROM THE DATA

Variables correlated		r_{xy}	P [1]
X	Y		
Loaf volume (Standard Basic)	Crude flour protein	+.7236	<.0001
Loaf volume (malt-phosphate-bromate)	Crude flour protein	+.7928	<.0001
Loaf volume (Standard Basic)	Wet crude gluten	+.7829	<.0001
Loaf volume (malt-phosphate-bromate)	Wet crude gluten	+.8866	<.0001
Loaf volume (Standard Basic)	Gluten protein in flour, %	+.7762	<.0001
Loaf volume (malt-phosphate-bromate)	Gluten protein in flour, %	+.8845	<.0001
Loaf volume (Standard Basic)	Protein removed by $MgSO_4$ (mg.)	+.2539	.2022
Loaf volume (malt-phosphate-bromate)	Protein removed by $MgSO_4$ (mg.)	+.2118	.2908
Loaf volume (Standard Basic)	Diastatic activity	−.3063	.1222
Loaf volume (malt-phosphate-bromate)	Diastatic activity	−.6982	.0001
Loaf volume (Standard Basic)	Starch resistivity	+.5786	.0002
Loaf volume (malt-phosphate-bromate)	Starch resistivity	+.7276	<.0001
Loaf volume (malt-phosphate-bromate)	Protein soluble in sodium salicylate, mg.	+.1457	.4668
Crude flour protein, %	Wet crude gluten	+.9522	<.0001
Crude flour protein, %	Flour ash	+.7422	<.0001
Crude flour protein, %	Protein removed by $MgSO_4$ (mg.)	+.3649	.0611
Crude flour protein, %	Protein soluble in sodium salicylate, mg.	+.0389	>.5485
Diastatic activity	Starch resistivity	−.7429	<.0001
Diastatic activity	Flour ash	+.1411	.4846

[1] P = probability of the observed correlation coefficient arising from uncorrelated material through errors of random sampling.

Tests of the significance of the differences between several of the correlation coefficients are listed in Table V. The transformation of the correlation coefficient r to the Z value was performed, as recommended by Fisher (1928), and the significance of the difference $Zr_1 - Zr_2$ was then evaluated in terms of the probability of the observed difference arising through sampling errors. The relative deviate, X^1, is found from the ratio of the difference between the Zr values and the standard deviation of the difference

$$\left(\sqrt{\frac{1}{N_1 - 3} + \frac{1}{N_2 - 3}}\right).$$

TABLE V

TESTS OF THE SIGNIFICANCE OF THE DIFFERENCE BETWEEN CORRELATION COEFFICIENTS

Correlation coefficient	Zr	$d/\sigma d$	X^1	P^1
.7928	1.0752			
.7236	0.9151	0.1601 / .2887	0.55	>.5485
d = (diff.)	0.1601			
.8866	1.4062			
.7829	1.0529	.3533 / .2887	1.22	.2229
d = (diff.)	0.3533			
.7829	1.0529			
.7236	0.9151	.1378 / .2887	0.48	>.5485
d = (diff.)	0.1378			
.8866	1.4062			
.7928	1.0752	.3310 / .2887	1.15	.2508
d = (diff.)	0.3310			
.7276	0.9230			
.5786	0.6604	.2626 / .2887	0.91	.3631
d = (diff.)	0.2626			

[1] P = probability of the observed difference arising through errors of random sampling.

There are no significant differences demonstrated among the five pairs of correlation coefficients shown in the table. Accordingly, the coefficients obtained between the malt-phosphate-bromate baking results and flour protein, and between malt-phosphate-bromate values and wet crude gluten, are not significantly higher than the corresponding values obtained with the Standard Basic results. Similarly, the correlation yielded by loaf volume and wet crude gluten is not significantly higher than the correlation between loaf volume and flour protein. No significant difference existed between the relations between starch resistivity and the loaf volumes registered by the two baking methods. It is therefore apparent that the use of an "improver" formula did not give a significant increase in information as compared with the Standard Basic Method in this set of millstream flours. This conclusion is in agreement with the results obtained by the senior author when working with Canadian hard red spring wheat mill-stream flours. A knowledge of flour protein yields as accurate information in regard to baking value, as registered by loaf volume, as a determination of wet crude gluten in these mill-stream flours.

As it appeared possible that the observed negative correlation between diastatic activity and loaf volume might be due in part at least to the influence of starch resistivity, partial correlation technique was employed to eliminate, first, the effect of diastatic activity upon the loaf volume—starch resistivity relationship. A positive relation-

ship between the two variables was obtained under these conditions. The effect of diastatic activity upon loaf volume when starch resistivity was held constant was also determined, and a relatively small negative correlation coefficient obtained. At least part of the relation between diastatic activity and loaf volume is apparently due to lowered starch resistance to diastatic action. This is especially true of flours coming from the germ reductions and the dust reel. These correlations are shown in Table VI.

TABLE VI

PARTIAL CORRELATIONS

Variables correlated	Variable held constant	Correlation coefficient
Starch resistivity and loaf volume (malt-phosphate-bromate)	Diastatic activity	+.4359
Diastatic activity and loaf volume (malt-phosphate-bromate)	Starch resistivity	−.3433

Table VII shows the method of predicting the percent of wet crude gluten in these flours from a knowledge of the flour protein content. The determination of crude gluten by the method of washing out the flour starch and other constituents as far as possible was considered to be very important at one time before the Kjeldahl test or its modifications came into general use for determining flour protein.

TABLE VII

PREDICTION OF PERCENT OF WET CRUDE GLUTEN FROM PERCENT OF CRUDE FLOUR PROTEIN

Linear regression coefficient
Percent of wet crude gluten$^{(y)}$ on percent of crude flour protein$^{(x)}$ $B_{xy} = 3.942$
Linear regression equation
Estimation of percent of wet crude gluten (y) from percent of crude flour protein (x) $y = -6.78 + 3.94x$ Standard error of estimate = 2.4%

Summary and Conclusions

A series of 27 mill-stream flours were analyzed and baked by two methods—the Standard Basic, and the malt-phosphate-bromate. Wet crude gluten was also washed from the flours, employing a 0.1% sodium phosphate solution whose pH was approximately 6.8. These glutens were then dispersed in 10% sodium salicylate solution and fractionated by the addition of a small quantity of $MgSO_4$. The data obtained appeared to justify the following conclusions.

1. The use of malt-phosphate-bromate does not yield significantly higher correlations with various quality factors than are obtained with the Standard Basic Method. It does, however, give greater differ-

entiation in regard to loaf volume between the samples, and brings out the potentialities of high gluten break flours in a striking manner. Its incorporation in methods for testing mill-stream flours produced from hard red spring wheat would therefore seem advisable.

2. The quantity of gluten protein removed from a sodium salicylate dispersion by a single small addition of $MgSO_4$ does not appear to vary appreciably from sample to sample in mill streams, regardless of the quality of the flour, and is not related to any appreciable degree with other factors of quality. The quantity of this constituent present in the gluten washed from flours originating in different portions of the wheat kernel therefore must be fairly constant. As it will vary in the flour according to the gluten content when calculated as percent of the flour, this fraction is highly and positively related to loaf volume.

3. The better quality flours appeared to have lower diastatic activity in these samples. A part of this effect at least was due to differences in the resistivity of the starch substrate to diastatic attack. Flours presumably containing germ particles exhibited high diastatic activity coupled with lower starch resistivity. It is probable that in one instance severe grinding had the effect of lowering starch resistance to injury to the starch granule, thus increasing its susceptibility to diastatic action.

4. Flour protein appeared to be as highly related as wet crude gluten percentage to loaf volume in this series of flours, thereby justifying the substitution of the Kjeldahl test for the gluten washing determination. Flour protein, as well as wet crude gluten and gluten protein, was high in the break flours. Flour protein tended to increase with ash content; that is, these two constituents appeared to be related to each other in their location in the wheat kernel.

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DIRECT CURRENT OF STABILIZED VOLTAGE FOR THE TAG-HEPPENSTALL MOISTURE METER AND OTHER LABORATORY INSTRUMENTS[1]

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(Received for publication January 26, 1937)

Numerous instruments requiring direct current at potentials above 40 volts are coming into routine laboratory use. In the past these have usually been supplied with current by dry batteries, but the tendency is now growing towards a rectified supply from the alternating current power lines. This is urged not only by the cost of periodic replacements of the batteries, but also by their gradual drop in voltage and their occasional failure at inconvenient times.

Various types of rectifier circuits and voltage control units have been developed for different radio uses, but if the most satisfactory and at the same time economical circuit is to be developed, the current requirements of the instrument to be supplied must first be studied. Certain photoelectric devices, electronic voltmeters and other instruments have a low and nearly constant current demand, which simplifies

[1] Contribution No. 54, Department of Milling Industry, Kansas Agricultural Experiment Station.

the requirements unless a very high degree of voltage constancy is required. If, however, the current drain varies widely, the rectifier design must be carefully planned or the voltage will drop seriously as the current increases.

The Tag-Heppenstall moisture meter has been found very useful for rapid determinations of moisture in grain where high accuracy is not essential. This meter requires a battery of 112.5 volts, and as used at this laboratory, replacements are necessary at least twice a year. Since the cost of these amounts to a considerable sum over a period of years, it seemed desirable to construct a rectifier to operate from the power line, and the study was continued to include an electronic voltage regulator for use with instruments requiring very stable voltage, or in cases where the voltage of the power supply fluctuates widely.

A study of the circuit of the Tag-Heppenstall meter shows that while it uses a current of only about 1 microampere when testing wheat of 8% moisture content, the current is nearly 15 milliamperes for wheat of about 24% moisture, the maximum for which the meter is designed. A modification of the meter recommended by Coleman and Fellows (1936) for increasing the range to include grain of higher moisture content increases the possible current consumption to 20 milliamperes at 90 volts, the limit set by the protective resistor of the meter. This full range is not usable as it corresponds to a short circuit of the electrodes, but the rectifier circuits described here have been required to deliver this current within the permissible voltage drop.

Accuracy Required

The permissible voltage variation may be estimated from the voltage drop allowed by the manufacturers in their instructions for testing batteries for satisfactory strength. The meter is standardized by switching to a tap marked *S* which connects to a standard resistor drawing about 1.5 milliamperes. A rheostat is then adjusted to bring the meter to a reading of 50. To check the condition of the batteries the switch is then changed to a tap marked *T*, connected with a standard resistor drawing about 13.5 milliamperes. Instructions are that if the meter reading drops below 48 on this tap the battery should be replaced. Assuming a voltage of 100, which is nearly the minimum useful voltage of the 75 cell battery used, this would apparently correspond to a voltage drop of 4 volts, but it actually corresponds to 6 volts since the standardizing resistors are so chosen that if the voltage does not drop, the meter reading rises to 51. Judging from this allowance, it was assumed that for a rectifier supply circuit a drop of

3 volts at the maximum current consumption would be well within the requirements for accuracy.

However, a more fundamental measure of the allowable voltage fluctuations may be obtained from the standard error of the instrument. The error of replication of the Tag-Heppenstall meter is very low, but since the electrical conductivity of wheat is not perfectly correlated with moisture content, the error of its prediction of moisture content is considerably higher. Cook, Hopkins, and Geddes (1934) report an observed standard error of prediction of 0.21% moisture in testing hard red spring wheats below 17% in moisture content using as a standard the vacuum oven with a drying period of 16 hours. For wheat from 17 to 24% in moisture content the observed standard error was 0.58% moisture, or nearly three times as much as with the drier wheat.

Coleman (1931) found an average deviation from air-oven tests of 0.17% as measured on 147 samples of hard red winter wheat, with 13 samples in error by 0.4% or more. However, Cook, Hopkins, and Geddes (1934) found the errors for the air-oven method greater than those for the Tag-Heppenstall meter, except for wheat moisture content above 17%.

Measurements made in the laboratories of the Milling Department of Kansas State College in comparison with the results given by the steam-shelf vacuum oven described by Anderson (1936) show that the error of the Tag-Heppenstall meter varies considerably from year to year. Excluding all samples which had been artificially wetted, since the error for these was found to be greater, the standard error was 0.29% moisture for the crop of 1935 and 0.49% for the crop of 1936. For the former the errors were about evenly divided, with a majority negative; for the latter year not only was a large majority of the errors positive, but the negative errors averaged as great as those of the preceding year. For these two years the samples not artificially wetted were all below 11% in moisture content, or lower than most of the samples tested by Cook, Hopkins, and Geddes (1934). The greater error is possible due to the fact that many varieties and different growing conditions were included.

From the accuracy of the results obtained from the meter in this laboratory it was judged that voltage fluctuations were unimportant if the resulting error in moisture indication was not over 0.1%, but considering the greater accuracy reported by other investigators, it was decided to limit the permissible variation to 0.05% moisture on all samples of 17% moisture or less. This appears very conservative, considering that for the vacuum-oven method, the most accurate moisture method available, Cook, Hopkins, and Geddes (1934) found a standard error of 0.118% moisture.

Since the accuracy of any grain moisture determinations, and particularly those made by the Tag-Heppenstall moisture meter, decreases rapidly at moisture contents above 17%, the theoretically permissible variation was allowed to increase gradually to 0.1% at 19% moisture content, 0.3% at 21% moisture, and 0.5% at the maximum reading of the instrument with the additional resistor recommended by Coleman and Fellows (1936).

Assuming these errors to be negligible, the permissible voltage fluctuation is plus or minus 4 volts or more at any point on the scale. This is a considerably more stringent requirement than that placed on the use of old batteries by the instructions of the manufacturer.

Simple Circuit

Probably the most easily assembled circuit falling within the above requirements is that shown in Figure 1 using the type 874 R.C.A.

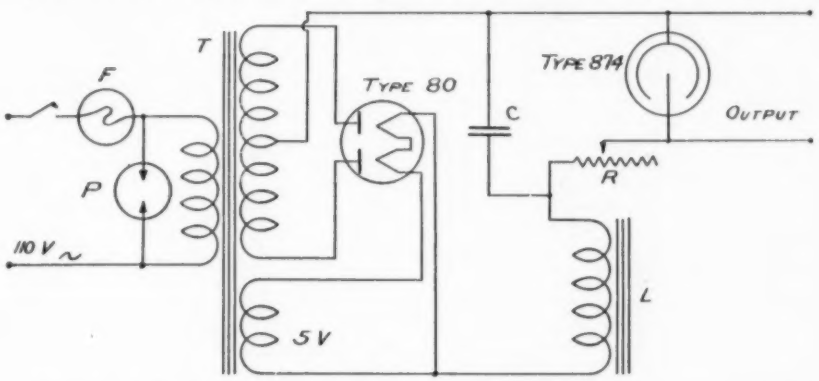

Figure 1. *C*—condenser, 8 mfd.; *F*—fuse, 1 ampere; *L*—choke, 11-henry, 65 milliamperes; *P*—neon lamp, ¼ watt; *R*—adjustable resistor, 6,000 ohms, 25 watts; *T*—power transformer, 50 milliamperes, about 340 volts each side of center tap.

gas discharge voltage regulator tube. The power transformer and choke may be inexpensive units as made for midget radio sets, but with current ratings of at least 50 milliamperes. When the circuit is assembled, a milliammeter should be connected in series with the type 874 tube and resistor *R* adjusted to give a current of 35 or 40 milliamperes when no current is being drawn from the output. Under these conditions voltage fluctuations up to 15% in the supply current will cause no appreciable change in the output voltage. A current drain of 20 milliamperes causes a voltage drop of about two volts, or only about half that permitted for the Tag-Heppenstall meter.

If for any reason it should be desirable, the amount of this voltage drop may be somewhat reduced by using a power transformer delivering about 130 volts each side of the center tap, and a type 83 or 82

tube instead of the type 80. The choke should then be of about 30 henries inductance, and the adjustable resistor may be rated at about 1,000 ohms, 10 watts. With this change, when the output voltage begins to drop there is a considerable increase in the current delivered by the rectifier. The output voltage fluctuation due to changes in the input voltage will be somewhat increased.

Adjustable Voltage

However, in this laboratory it was desired to experiment with voltages as low as 25 volts for wheats of high moisture content and as high as 160 volts for wheats of low moisture content. Obviously the circuit just described would not furnish such voltages, so the circuit shown in Figure 2 was used. It is apparent that the output

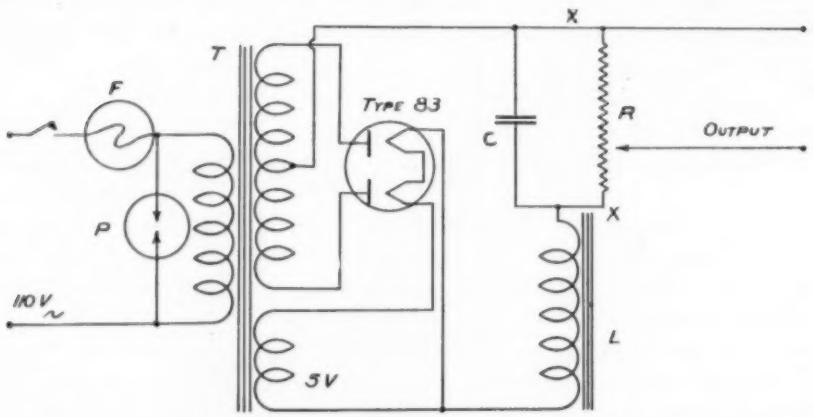

Figure 2. *C*—condenser, 8 to 16 mfd.; *F*—fuse, 1 ampere; *L*—swinging choke, 8–30 henry at 250–0 milliamperes; *P*—neon lamp, ¼ watt; *R*—adjustable resistor, 1,000 ohms, 200 watts; *T*—power transformer, 225 milliamperes, 200 volts each side of center tap; *X*, *X*—points for substitution of output shown in Figure 2*A*.

voltage will drop as the current drain increases, and the following precautions should be observed to minimize this effect: The power transformer should operate at fairly near magnetic saturation, and should deliver a voltage as low as will supply the required output voltage. The rectifier tube should preferably be of the mercury vapor type, though the 83 *V* tube gives satisfactory results. A choke of fairly high inductance should form the filter input. The current flowing continuously through voltage divider *R* should be as high as practicable.

As assembled from readily available and inexpensive radio parts according to the specifications given, a voltage drop of 3 volts is observed at a drain of 20 milliamperes, the maximum consumption of the Tag-Heppenstall meter. When tested with the moisture meter

itself according to the method described for testing batteries, the reading drops only ⅓ scale division when the meter switch is changed from tap S to tap T, instead of 2 scale divisions, the limit set by the manufacturers.

This circuit does not compensate for line voltage fluctuations beyond the regulation characteristics of the power transformer used. This has been satisfactory here as the line voltage is relatively constant, and four-volts fluctuation in input at 220 volts causes only one volt change in output at 100 volts. If occasional voltage fluctuations of considerable magnitude must be guarded against, the addition of a voltmeter, V, and a readily adjustable power rheostat, R_2, as shown in Figure 2*A*, forms a satisfactory solution. If the moisture meter is to be operated only at normal voltage, the power transformer may be selected to deliver about 130 volts each side of the center tap, R_1 may

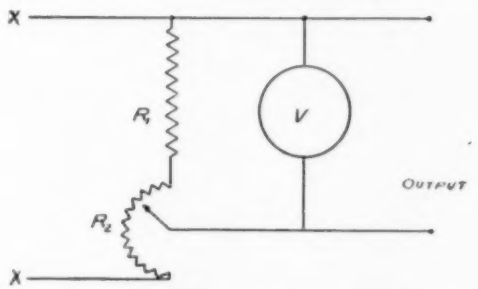

Figure 2*A*. R_1, R_2—resistor and power rheostat (potentiometer connection) specifications as required by the power transformer used; V—voltmeter, full scale reading equal to maximum voltage to be drawn from circuit.

be rated at 500 ohms, 100 watts, and R_2 at 100 ohms, 25 watts. V may be an inexpensive meter of 100 volt range; its accuracy need not be high but the scale should be long enough that fluctuations over two or three volts may be readily seen and corrected by a change in R_2. This adjustment may easily be made while the moisture meter is in operation if the motor-driven model is used, and as previously noted, fluctuations are insignificant unless they exceed four volts. With the power transformer delivering only 130 volts, the voltage drop at the meter at its maximum current consumption is only two volts, too little to require readjustment of the rheostat.

Electronic Voltage Regulator

In order to furnish a power supply of highly accurate voltage for comparative testing, the circuit shown in Figure 3 was designed and assembled. This is a modification of a circuit designed by R.C.A. for their type 874 voltage regulator tube. The latter circuit could not

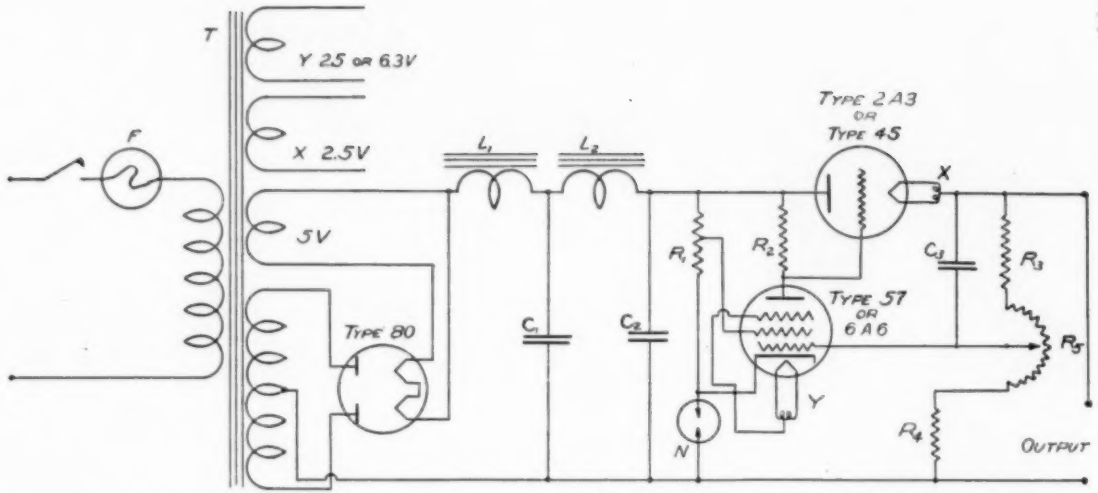

Figure 3. C_1, C_2—condensers, 8 mfd.; C_3—paper condenser, 2 mfd. or more; F—fuse, 1 ampere; L_1, L_2—chokes, 11-henry, 65 milliamperes; N—neon glow lamp, 2-watt, with resistor removed from base; R_1—adjustable resistor, 25,000 ohms, 10 watts; R_2—resistor, 3 megohms, 1 watt; R_3, R_4—resistors, 10,000 ohms, 5 watts; R_5—potentiometer, linear wire-wound volume control, 10,000 ohms; T—power transformer, 3 filament windings, high voltage 50 milliamperes, 350 volts each side of center tap; X—2.5-volt filament winding for power tube, center tapped, or with 20-ohm center-tapped resistor; Y—2.5-volt or 6.3-volt filament winding for control tube, center tapped, or with 30-ohm center-tapped resistor.

be used since the voltage of the regulator tube plus the necessary bias for the power tube gives an output voltage too high for the Tag-Heppenstall meter. The ordinary 2-watt neon glow lamp with protective resistance removed from the base gives very satisfactory results as a regulator, though its voltage varies more than that of the type 874 for a given change in operating current. The screen of the type 57 tube is ordinarily connected to a point on R_1 with a potential about 80 volts positive to the cathode, the exact setting depending on individual circuit characteristics. If the output voltage increases with increased current drain, lower the screen voltage, and *vice versa*.

The specifications given are for inexpensive parts satisfactory for delivering up to 20 milliamperes at about 100 volts. Table I gives the maximum range of variation of input voltage over which output voltage will be accurately regulated for any current drain from 0 to 20 milliamperes: The output is adjusted to the voltage given, or any

TABLE I

PERMISSIBLE INPUT VOLTAGE FLUCTUATION FOR ACCURATE VOLTAGE REGULATION AT ANY CURRENT DRAIN, 0 TO 20 MILLIAMPERES

Input voltage, type 2A3 tube	Input voltage, type 45 tube	Output voltage adjusted to
82 to 130	90 to 120	100 volts
85 to 150	90 to 125	105 "
87 to 150		110 "

intermediate value, by the potentiometer R_5. Higher voltages are available at current drain lower than 20 milliamperes. Except at the extremes of this range, output voltage fluctuations should not be discernible on a 150-volt meter with a 5-inch scale. This has been found satisfactory as current source for a photoelectric colorimeter and for a high impedance vacuum-tube voltmeter.

If currents much larger than 20 milliamperes are to be drawn, or if higher output voltages are desired, the power transformer should deliver a higher voltage, and the transformer, rectifier tube, and chokes should have current capacities with ample reserve above the expected drain. It would be well to increase the resistance of the bleeder system, R_3–R_4–R_5, and in any case R_1 should be so chosen as to pass approximately 10 milliamperes through N. The type 45 tube will not be satisfactory for currents above about 30 milliamperes, but the 2A3 will easily deliver 50 milliamperes, and for higher currents the circuit may be adapted to use the 6L6 tube, or two of them in parallel.

In Figure 3 the regulating glow lamp *N* may be so mounted as to serve as a pilot lamp, therefore no other pilot has been shown. In Figures 1 and 2 an alternative to the neon pilot lamp is a 6.3-volt, 0.15-ampere radio panel light connected across the 5-volt filament winding of the power transformer. At this reduced voltage the bulb will draw very little current and last almost indefinitely.

Summary

The current requirements of the Tag-Heppenstall moisture meter are studied and two rectifier circuits are shown which fulfill them. The second circuit permits adjustment of its output voltage over a wide range. A third circuit is shown incorporating an electronic regulator which will maintain the output voltage constant within 0.1 volt or less over a wide range of input voltages at any current drain within the capacity of the power tube.

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THE RELATIONSHIP BETWEEN DIASTATIC ACTIVITY (MALTOSE FIGURE) AND "GASSING POWER" OF EXPERIMENTALLY MILLED FLOURS FROM SOME AUSTRALIAN WHEATS

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(Received for publication December 22, 1937)

Introduction

The importance of the diastatic activity of flour is too well recognised to need stressing here. Commercially, increasing control is being carried out both by the addition of malted wheat flour and by careful wheat blending. This necessitates accurate and, if possible, quick methods of analysis. Several methods are available for the determination of diastatic activity and gassing power from the point of routine mill control, but the selection of any one is a matter of choice, or circumstance, and opinions still differ with regard to their relative merits.

In this laboratory the routine method used for the determination of diastatic activity or maltose figure is Kent-Jones' modification of Rumsey's method while the Brabender Fermentograph is used for the determination of "gassing power." However, it was found over a period of 18 months that the gas production of a dough, as judged by the diastatic activity so determined, frequently failed to correspond with that shown by the Fermentograph.

This experiment was, therefore, carried out with the object of examining the relationship between the above two methods, though later the determination of diastatic activity by the Blish-Sandstedt method was included for comparative purposes.

Methods

1. Diastatic Activity

(*a*) *Kent-Jones' modification of Rumsey's method.* This method, as described by Kent-Jones (1927), was followed except that 15 instead of 30 g. of flour were used per test, and water, acid, *etc.* were reduced in proportion. Briefly, the method consists of digesting an unbuffered flour suspension for 1 hour at 27° C., then clarifying and preventing further diastatic activity by means of sodium tungstate solution and

sulphuric acid. The suspension is next filtered and the clear filtrate is titrated against Fehling's solution, using methylene blue as an internal indicator.

(*b*) *Blish-Sandstedt method.* This method is widely used for routine work and was tentatively recommended as a standard method by the conference of (Government) cereal chemists held in Sydney, December 1936. According to Davis and collaborators (1937) the method is about as satisfactory as the Schoorl and Bertrand methods for measuring reducing sugars after diastasis. It was, therefore, included in this experiment for comparative purposes and was used as set out in the A.A.C.C. *Cereal Laboratory Methods* (1935). (See also Davis and Worley, 1934.) In this method buffered flour suspensions are digested at 30° C. for 1 hour, after which the diastatic activity is inhibited as before with sodium tungstate solution and sulphuric acid. An aliquot of the filtrate is next treated with excess standard potassium ferricyanide, the unreduced ferricyanide being determined by titration with standard sodium thiosulphate.

2. "Gassing Power"

The gassing power was measured by the Brabender Fermentograph in the usual way. That is, the flour, yeast, salt and water dough was made in the Farinograph mixing bowl by mixing until the maximum dough development had been reached at the 600 unit line, usually about 4 minutes. Four hundred grams of the dough were then quickly weighed off, transferred to the Fermentograph balloon, fitted to the recording device and immersed in the water bath at 30° C. By knocking back the dough at the end of each hour and allowing the carbon dioxide produced during fermentation to escape, it was possible to measure the hourly rate of production of CO_2. Fermentation was continued until the rate had fallen to a low figure compared with the maximum or until 7 hours had elapsed.

The only question that arises in this test is the selection of the yeast and salt quantities. Several formulae were tried but the one finally adopted was that recommended by Brabender, *i.e.* $2\frac{2}{3}\%$ yeast and $1\frac{2}{3}\%$ salt, with which the gas produced approximately corresponds to that formed in a 3-hour dough (4 hours from dough to oven).

Selection and Preparation of Samples

Since we are primarily concerned with controlling the diastatic activity and other properties of the flour by means of wheat blending, the consideration of experimental mill flours is of paramount importance. The flours used in this test were, therefore, experimentally milled flours and it was decided to examine those from some wheat

varieties known to differ in their diastatic activity and also some about which a certain amount of controversy still obtains.

Ten-pound samples of wheat (1936–37 season) were supplied by various official organisations and were definitely *bona fide* samples of the stated variety. They are listed below together with the comments received with them.

Laboratory number	Sample	Source of supply and comments
1	Ranee 4H. Vic.	Dookie Agricultural College—experimental area.
2	Dundee. Vic.	
3	Nabawa. N.S.W.	New England Experiment Farm Glenn Innes. This sample is not from one plot, but from 5 replications.
4	Baringa. N.S.W.	Agricultural Department N.S.W. "Each sample is made up of good typical samples from a number of districts and may be taken as quite representative of the varieties."
5	Pusa. N.S.W.	
6	Ranee 4H. S.A.	Turretfield Seed Farm
7	Ford. S.A.	
8	Nabawa. S.A.	
9	"Selection 28." S.A.	
10	Ford. S.A.	Roseworthy Agricultural College
11	Ranee. S.A.	
12	Nabawa. W.A.	Wongan Hills Research Station. "Soil is very sandy and gravelly and hence wheats are low in protein. In the 1935–36 season 30.7% of the total acreage sown to wheat in Western Australia was Bencubbin and 8% sown to Nabawa."
13	Bencubbin. W.A.	

After examination 3,000 g. of each sample were conditioned and then ground in the Brabender experimental mill. Conditioning varied slightly with the wheat, the harder wheats such as the Pusa sample being given a little more water than the others, but in each case the samples were conditioned overnight with sufficient water to bring their moisture content to 15%. Then in most cases, ½ to 2% of water was added to the sample ½ to 1 hour before grinding. The variation in conditioning was made in order to produce within as narrow limits as possible flours with a moisture content of 15.0% and an extraction of approximately 58%. The flours were stored in air-tight tins throughout the experiment.

The flours were subjected to gluten, protein, moisture and ash tests besides the diastatic activity determinations, and their water absorption and strength curves were determined by the Farinograph. In order that a definite idea of the samples under consideration may

be obtained, the results of these analyses are given in Table I, where the samples have been tabulated in order of their strength as judged from their analysis and their Farinograph curve.

It is naturally difficult to compare such widely differing varieties as Baringa and Ranee, as has been done above, since the question as

TABLE I

SAMPLES ARRANGED IN ORDER OF STRENGTH (ANALYTICAL FIGURES BASED ON FLOUR OF 15.0% MOISTURE CONTENT)

Sample	Extraction	Ash	Water absorption	Dry gluten	Protein (N×5.7)	Comments
	%	%	%	%	%	
Pusa. N.S.W.	53.9	0.716	68.0	16.8	13.08	Strong, hard wheat. Very good sample.
Ford. S.A. (R)	61.7	0.518	58.0	15.8	11.54	Medium strength wheat, stability not so good as with Pusa sample.
Baringa. N.S.W.	55.2	0.756	71.3	18.4	14.19	Medium strength. Hard vitreous grains, stability low for this protein content.
Dundee, Vic.	59.3	0.585	66.0	11.8	9.56	Medium—nearly 50% mottled grains.
Nabawa. N.S.W. (G.I.)	61.8	0.440	59.6	10.1	8.61	Medium—very good stability for such a low-protein figure.
Ranee. S.A. (R)	63.6	0.442	60.1	11.7	9.43	Weak but good. Excellent sample of Ranee wheat.
Ford. S.A. (T)	58.2	0.430	54.5	10.4	8.95	Weak but good. Very stable but does not compare with sample from Roseworthy.
Ranee 4H. S.A. (T)	58.1	0.464	59.2	10.1	8.60	Weak. Grains very slightly bleached.
Nabawa. S.A. (T)	55.3	0.500	52.0	8.4	7.34	Weak, but very good stability.
Bencubbin. W.A.	58.7	0.524	57.8	10.6	8.68	Weak. Grains extremely long and plump.
Ranee 4H. Vic.	56.0	0.436	59.3	10.0	8.55	Weak. Sample had a good appearance.
"Selection 28." S.A. (T)	54.1	0.540	55.0	9.4	8.14	Weak—average sample.
Nabawa. W.A.	63.3	0.522	59.0	7.0	7.01	Very poor with practically no stability. Milled very well.

to whether one is or is not "better" than the other depends to a large extent on the individual requirements of the miller. However, considering samples of the one variety it may be seen that they fall in the following order in regard to "strength:"

Ranee	Nabawa	Ford
1. Roseworthy, S.A.	Glenn Innes, N.S.W.	Roseworthy, S.A.
2. Turretfield, S.A.	Turretfield, S.A.	Turretfield, S.A.
3. Dookie, Vic.	Tongan Hills, W.A.	
4. "Selection 28." Turretfield, S.A.		

The "Selection 28" sample has been placed with the Ranee samples because of the similarity of their characteristics.

It should be noted here that the samples of Bencubbin and Nabawa wheats from Western Australia are not representative as regards "strength" of the wheat grown in that State.

Relationship between Experimentally Milled Flours from the Brabender Mill and Corresponding Commercial Flours

Unlike the flour produced by the majority of experimental mills, that from the Brabender mill has a diastatic activity (as measured by above methods) considerably higher than the corresponding commercial flour. The relationship varies with the type of wheat ground, the difference being greater with harder than with soft wheats. On average, with soft wheats, such as the Ranee samples mentioned above, the diastatic activity as measured by the Kent-Jones method is from 20 to 40 mg. per 10 g. flour lower in the commercial than in the corresponding experimental flour. With harder wheats, such as Dundee, the difference may vary from 100 to 140 mg. per 10 g. of flour. The differences in gassing power as determined by the Fermentograph are however, not so marked, as may be seen from Table II, in which details of some typical results are given.

Samples *A* and *B* are soft wheat blends, while *C* and *D* are hard wheat blends. In each case the production of carbon dioxide from the commercially milled flour follows fairly closely that produced

TABLE II

DIASTATIC ACTIVITY AND GASSING POWER OF CORRESPONDING EXPERIMENTALLY AND COMMERCIALLY MILLED FLOURS

Fermentograph Gassing Power—C.Cs. CO_2 Produced per Hour

Sample	*A*		*B*		*C*		*D*	
Flour	Experimental	Commercial	Experimental	Commercial	Experimental	Commercial	Experimental	Commercial
1st hour	330	340	410	400	270	290	330	320
2nd hour	460	410	470	460	420	370	460	420
3rd hour	540	490	605	600	410	420	540	470
4th hour	570	490	620	580	470	450	530	460
5th hour	490	420	520	505	400	380	470	400
6th hour	380	345	310	315	350	340	420	360
7th hour	230	245	190	190	320	270	360	265
5-hour total	2390	2150	2625	2545	1970	1910	2330	2070
7-hour total	3000	2740	3125	2750	2640	2520	3110	2695
Diastatic activity—mg. maltose per 10 g. flour (Kent-Jones method)	237	209	248	218	384	248	426	287

from the experimentally milled flour, but is nearly always just a little below in value. As the samples are blends they can not be strictly compared with the previous results which are from definite varietal samples, but serve to indicate the characteristics of the commercial flour corresponding to these samples.

From a limited number of analyses it would seem that the original sucrose value of commercially milled flours from soft wheats is also below that of corresponding flours milled on the Brabender mill by approximately 30 mg. per 10 g. of flour. This figure is only tentative and may be altered after more analyses have been carried out. No determinations have yet been made on commercial flours from hard wheats.

Results of Diastatic Activity Measurements

Two or more determinations of diastatic activity were made on each sample both with the Kent-Jones and Blish-Sandstedt methods. The average results are shown in Table III together with the total

TABLE III

DIASTATIC ACTIVITY AND GASSING POWER

Sample number	Mg. maltose per 10 g. flour		C.c. of CO_2 (Fermentograph method)	
	Kent-Jones method	Blish-Sandstedt method	After 5 hours	After 7 hours
1	170	148	1970	
2	449	383	2040	2770
3	227	208	1810	
4	366	300	2590	3140
5	351	293	2590	3460
6	197	220	1990	2680
7	112	107	1490	
8	154	157	1910	
9	167	198	1970	2590
10	111	115	1670	
11	185	220	2250	2805
12	348	458	1890	2460
13	188	198	2320	2860

amount of carbon dioxide produced in five and in seven hours in the Fermentograph test. The results are based on flour at 15.0% moisture content since it was approximately at this moisture content that the tests were carried out.

In Table IV the samples have been listed in the order of their diastatic activity, while in Table V are given the results of analysing standard sugar solutions by the Kent-Jones and Blish-Sandstedt methods.

TABLE IV

ORDER OF DIASTATIC ACTIVITY AND GASSING POWER OF SAMPLES

Order	Diastatic activity [1]		Gassing power	
	By Kent-Jones method	By Blish-Sandstedt method	By Fermentograph C.c. of CO_2	
			5-hour total	7-hour total
1st	No. 2. (449)	No. 12. (458)	No. 4. (2590)	No. 5. (3460)
2nd	4. (366)	2. (383)	5.	4. (3140)
3rd	5. (351)	4. (300)	13. (2320)	13. (2860)
4th	12. (348)	5. (293)	11. (2250)	11. (2805)
5th	3. (227)	6.	2. (2040)	2. (2770)
6th	6. (197)	11. (220)	6. (1990)	6. (2680)
7th	13. (188)	3. (208)	1.	9. (2590)
8th	11. (185)	13.	9. (1970)	12. (2460)
9th	1. (170)	9. (198)	8. (1910)	
10th	9. (167)	8. (157)	12. (1890)	
11th	8. (154)	1. (148)	3. (1810)	
12th	7. (112)	10. (115)	10. (1670)	
13th	10. (111)	7. (107)	7. (1490)	

[1] Milligrams of maltose per 10 g. of flour.

TABLE V

COMPARATIVE TESTS OF STRENGTH OF MALTOSE SOLUTIONS

(Maltose values expressed as milligrams maltose per 10 g. flour. All results are the mean of two or more determinations.)

Theoretical maltose value	Kent-Jones method		Blish-Sandstedt method	
	Mg. of maltose	% of theoretical	Mg. of maltose	% of theoretical
150 (Buffered solution)	197.5	98.8	141	94.1
200 (Buffered solution)			182	91.0
350 (Buffered solution)			318	90.8
150 (Unbuffered solution)			126	84.0
200 (Unbuffered solution)	199.7	99.9	168	84.0
296 (Unbuffered solution)	296	100.0	256	86.5
400 (Unbuffered solution)	403.5	100.9	340	85.0

The buffered solutions used in the above analyses were made as follows: 2.5 g. of B.D.H. maltose were dissolved in water to give a 1.0% solution. To 50 ml. of this solution there were added 180 ml. of buffer solution, 10 ml. of 10% H_2SO_4 and 10 ml. of 12% tungstate solution.

The other solutions were made up in a similar manner, proportional amounts of buffer and acid solutions, *etc.*, being used. They were then tested as for flour extracts. (See Davis, 1937.)

The flours were also tested for original sucrose content by inverting an aliquot of the water extract with hydrochloric acid, diluting to half the original concentration and then titrating against Fehling's solution (Kent-Jones, 1927).

The results of the sucrose estimations are given in Table VI.

TABLE VI

SUCROSE IN FLOURS (MILLIGRAMS PER 10 G. FLOUR)

Sample	Sucrose (mean of determinations)
1. Ranee, Vic.	269
2. Dundee, Vic.	218
3. Nabawa, N.S.W.	229
4. Baringa, N.S.W.	269
5. Pusa, N.S.W.	222
6. Ranee, S.A.	317
7. Ford, S.A.	198
8. Nabawa, S.A.	249
9. "Selection 28," S.A.	268
10. Ford, S.A.	170
11. Ranee, S.A.	272
12. Nabawa, W.A.	283
13. Bencubbin, W.A.	317

In order more easily to relate the gassing power with maltose and sucrose contents, the hourly gas production for each sample has been given in Table VII together with these figures.

TABLE VII

DIASTATIC ACTIVITY, SUCROSE CONTENT AND CO_2 PRODUCTION PER HOUR

Sample	Diastatic activity Kent-Jones method	Original sucrose	CO_2 produced—c.c. per hour							Total CO_2 (c.c.) produced	
	(Milligrams per 10 g. flour)		1.	2.	3.	4.	5.	6.	7.	After 5 hours	After 7 hours
1. Ranee	170	269	370	400	460	470	270	150	—	1970	
2. Dundee	449	218	260	340	460	520	460	390	340	2040	2770
3. Nabawa	227	229	260	400	470	450	230	—	—	1810	
4. Baringa	366	269	240	440	580	650	680	370	180	2590	3140
5. Pusa	351	222	230	400	600	680	680	560	310	2590	3460
6. Ranee	197	317	300	420	440	440	390	380	300	1990	2680
7. Ford	112	198	300	350	460	260	120	—	—	1490	
8. Nabawa	154	249	350	370	480	440	310	190	—	1910	
9. "Selection 28"	167	268	350	430	460	380	350	340	280	1970	2590
10. Ford	111	170	320	380	480	270	120	—	—	1670	
11. Ranee	185	272	320	405	520	550	460	350	205	2250	2805
12. Nabawa	348	283	370	410	410	340	340	290	280	1890	2460
13. Bencubbin	188	317	390	560	500	470	400	330	210	2320	2860

Discussion

From Table III it will be seen that there is not a constant relationship between the two methods used for the determination of the reducing sugar after diastasis, for the Blish-Sandstedt figures range from 66 mg. (Nos. 2 and 4) below, to 110 mg. (No. 12) above those obtained by the Kent-Jones method. Neither method shows a good correlation with the Fermentograph results (see Table IV) which, from both practical and theoretical considerations, are the more likely results to be obtained in baking practice.

Assuming therefore that the order based on Fermentograph results in which the samples are placed is correct, it will be seen from Table IV, or perhaps more easily from Table VIII, that both methods used

TABLE VIII

ORDER OF DIASTATIC ACTIVITY AND GASSING POWER OF SAMPLES

(*Table IV Showing Actual Varieties*)

Order based on diastatic activity		Order based on gassing power
Kent-Jones method	Blish-Sandstedt method	Fermentograph (5-hour total)
No. 2. Dundee, Vic.	No. 12. Nabawa, W.A.	No. 4. Baringa, N.S.W. }
4. Baringa, N.S.W.	2. Dundee, Vic.	5. Pusa, N.S.W. }
5. Pusa, N.S.W.	4. Baringa, N.S.W.	13. Bencubbin, W.A.
12. Nabawa, W.A.	5. Pusa, N.S.W.	11. Ranee, S.A. (R)
3. Nabawa, N.S.W.	11. Ranee, S.A. (R) }	2. Dundee, Vic.
6. Ranee, S.A. (T)	6. Ranee, S.A. (T) }	6. Ranee, S.A. (T)
13. Bencubbin, W.A.	3. Nabawa, N.S.W.	1. Ranee, Vic. }
11. Ranee, S.A. (R)	9. Selection 28, S.A. }	9. Selection 28, S.A. }
1. Ranee, Vic.	13. Bencubbin, W.A. }	8. Nabawa, S.A. (T)
9. Selection 28, S.A.	8. Nabawa, S.A. (T)	12. Nabawa, W.A.
8. Nabawa, S.A. (T)	1. Ranee, Vic.	3. Nabawa, N.S.W.
7. Ford, S.A. (T)	10. Ford, S.A. (R)	10. Ford, S.A. (R)
10. Ford, S.A. (R)	7. Ford, S.A. (T)	7. Ford, S.A. (T)

for the determination of reducing sugars after diastasis show about the same degree of misplacements as compared with the Fermentograph results (5-hour totals). The discrepancies between actual gassing power and expected gassing power as judged from the diastatic activity measurements are not uniform, the actual gassing power being sometimes above (*e.g.*, Bencubbin W.A. sample) and sometimes below (*e.g.*, Nabawa W.A. and Dundee Vic. samples) the values expected. This variation cannot be attributed to grinding effects on wheats of different physical characteristics. Rather does it illustrate the conclusions reached by Sandstedt, Blish, Mecham, and Bode (1937)

that some flours probably contain a small amount of "available" starch together with an enzyme or "activator" that is responsible for rendering the raw starch available to the sugar-producing enzyme, beta-amylase. The content of this particular enzyme or "activator" varies from flour to flour, apparently being greater in the hard than in the soft wheat flours. In the diastatic activity test the length of time involved is not sufficient for this enzyme to become significant, *i.e.*, sugar is still being produced from the available starch, consequently there is no indication of its activity or concentration as there is in the gassing power test.

From the results given in Table V, obtained by analysing standard sugar solutions by the two different methods under consideration, it will readily be seen that the Kent-Jones method has a lower experimental error. Its error only varies 1.0% (from 99.9 to 100.9%) over a range of maltose values from 150 to 400 mg. per 10 g. of flour while the Blish-Sandstedt method shows a variation in the experimental error of 3.3% (90.8 to 94.1%) over a range of 150 to 350 mg. maltose per 10 g. flour, the error increasing with increasing concentration of maltose. The results also demonstrate the need for correct buffering of the sugar solutions when the latter method is employed.

When it was seen that the correlation between diastatic activity and gassing power was rather irregular, determinations of original sucrose were carried out on the test samples.

The results (see Table VII) varied rather widely and were mainly remarkable for their high value. They show how it is possible for a flour with a low diastatic activity figure, such as sample 13, to produce when fermented in a dough more carbon dioxide than would be expected from a consideration of its diastatic activity alone. On the other hand, sample 12 has both a high diastatic activity and initial sucrose content and yet only ranks 10th on its gassing power (5-hour total) thus indicating that a third factor is involved in gas production. This factor we presume to be the enzyme or "activator" discussed above.

The grouping of the samples according to the Fermentograph test (gas produced per 5 hours) is noteworthy because although only a limited number of samples have been considered, they have, apart from one exception (No. 2 Dundee), fallen into varietal groups (Table VIII).

The amount of gas produced by the samples in 5 hours by the Fermentograph method has been used for comparative purposes because it is practically more significant than the 7-hour gas-production figure since the yeast-salt ratio was chosen for a 5-hour dough.

Summary and Conclusions

A comparison has been made, from the point of view of routine mill control, of the relationship between diastatic activity, as measured by the Kent-Jones modification of Rumsey's method and by the Blish-Sandstedt method, and gassing power, using the Brabender Fermentograph, of flours from different Australian varieties of wheats which have been milled on a Brabender experimental mill. Although the number of wheat samples considered here is limited the analytical results, in the author's opinion, are fairly indicative of the particular varieties examined unless otherwise stated. Sucrose estimations have been made and a comparison has been drawn between flours from wheats milled on the Brabender experimental mill and their corresponding commercial flours.

The conclusions are as follows:

Measurement of diastatic activity. A poor correlation was obtained between the Blish-Sandstedt and Kent-Jones methods for measuring reducing sugars after diastasis, the difference between them varying from positive to negative in an inexplicable manner.

The Kent-Jones method has a lower experimental error and is more suited to routine work than the Blish-Sandstedt method since it is faster, more economical in chemicals, and there is less possibility for mistakes in technique.

Correlation between diastatic activity and gassing power. The correlation between diastatic activity and gassing power is on the whole rather poor though it is slightly better with the Blish-Sandstedt than with the Kent-Jones method, but not sufficiently so to outweigh the advantages outlined above of the Kent-Jones method.

Since diastatic activity measurements do not account for all the factors which influence gas production, the results so obtained must necessarily be less informative than methods which actually measure gas production.

The results obtained in this experiment show that whereas a diastatic activity measurement may give some indication of the probable gassing power of a flour, it may frequently be misleading and should always be supplemented by a gassing power test in the case of flours from "unknown" wheat samples.

The main use of diastatic activity measurements is in mill-control work when the type of flour under consideration varies only within narrow limits.

The Brabender Fermentograph affords a logical, easy and accurate method for the measurement of gassing power.

Varieties considered. The number of samples examined is too small to make definite statements regarding the gassing power of different varieties but the indications are definitely that the gassing power is a varietal characteristic which, unlike "strength," tends to be of the same order irrespective of the district in which the sample is grown, *e.g.* Ranee, Nabawa and Ford samples. The hard wheats, Pusa and Baringa, are most vigorous in gassing power while the Ford variety is undoubtedly the poorest. The Ford samples had the lowest diastatic activity as determined by both the Blish-Sandstedt and the Kent-Jones methods, the lowest sucrose content and the lowest gassing power.

Experimental and corresponding commercial flours. Flours from wheats milled on the Brabender experimental mill show higher diastatic activity figures and higher gassing power than corresponding commercial flours though the Fermentographs show less difference than do the diastatic activity figures. Typical relationships have been set out in Table II.

Acknowledgment

The author wishes to express his appreciation to the donors of the samples used in this experiment, and to W. S. Kimpton & Sons for permission to publish this report.

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STUDIES ON THE STORAGE OF WHEATEN FLOUR: III. CHANGES IN THE FLORA AND THE FATS AND THE INFLUENCE OF THESE CHANGES ON GLUTEN CHARACTER

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(Received for publication December 22, 1937)

A continuation of the previous investigations on the changes that take place in flours during storage carried out in these laboratories (Fisher, Halton, and Carter, 1937) has fully confirmed earlier results. It was shown in the earlier investigations that marked chemical changes occur in flour kept under storage conditions over a prolonged period of time. It was suggested that perhaps biological agencies such as bacteria and fungi, and possibly mites, may have been responsible for them. The earlier investigations were entirely chemical in nature and the determinations made then have been repeated, together with certain additional analyses. At the same time a biological examination has been carried out to ascertain whether the bacterial and fungal flora of flour show any marked changes in nature and number during storage, which could be correlated with the chemical changes that have been observed.

The only systematic bacteriological investigation of flour of which we are aware is that carried out by Kent-Jones and Amos (1930). It was shown by these investigators that the bacterial numbers of different flours vary enormously. For example, it was found by them that in a patent flour the bacterial numbers may be as high as 70,000 per gram, while in low-grade flours still higher figures were recorded, up to 600,000 per gram, whereas straight runs were found to occupy a somewhat intermediate position, up to 120,000 per gram. It was further discovered by Kent-Jones and Amos that on storage the bacterial numbers decreased. Flours stored for as short a period as 26 days showed a fall of over 50% over the initial values, while after storage for 70 days, the value per gram of flour fell even further.

In the present investigation, the preliminary storage experiments were carried out with two different low-grade flours, an English (bot-

[1] The author wishes to express thanks to E. A. Fisher (Director of Research) for suggesting this investigation and for his interest and valuable suggestions while the work was in progress.

tom 40%) and Manitoba (bottom 50%). These were stored in tins at two different moisture contents. The English flour had an initial moisture content of 16.48% and the Manitoba 14.56%. A portion of each of these flours was moistened to approximately 18%. Samples of the flours were removed at fortnightly intervals for the various routine analyses and bacterial and fungal counts.

For the bacterial counts, and also later, for the fungal counts, the technique originally devised by Kent-Jones and Amos proved to be very suitable. In this method 100 c.c. of a 0.5% sterile saline solution are placed in a wide-necked sterile glass bottle and 10 g. of freshly ignited sand added. Ten grams of the sample of flour to be investigated are accurately weighed out in a sterile dish and added to the saline-sand mixture and the whole shaken for 2 minutes. The mixture is allowed to settle and 5 c.c. are pipetted into 45 c.c. of sterile saline solution which is also contained in a wide-necked sterile glass bottle. This is now shaken for 1 minute, and after the mixture has settled a 5 c.c. aliquot is pipetted into a further 45 c.c. of sterile saline solution. Finally, 1-c.c. lots are pipetted out of this last dilution into sterile petri dishes for the plating out. This degree of dilution was found sufficient in the great majority of cases for the bacterial and fungal numbers encountered in the different flours used in this work.

In the preliminary stages of the investigation, nutrient beef or Lemco agar and gelatin were used for the plating out. Later, however, both beef and Lemco agar and gelatin were abandoned and the synthetic medium devised by Thornton (1922) was employed instead. The Thornton medium has the following composition:

K_2HPO_4	1.0	g.
$MgSO_4 \cdot 7H_2O$	0.2	g.
$CaCl_2$	0.1	g.
NaCl	0.1	g.
$FeCl_3$	0.002	g.
KNO_3	0.5	g.
Asparagine	0.5	g.
Mannitol	1.0	g.
Agar	15.0	g.
Water	1000	c.c.

To make up the medium, phosphate, nitrate and asparagine are dissolved in the same sample of water in the order named, and the $MgSO_4$, $CaCl_2$, NaCl and $FeCl_3$ in separate quantities of water which are then added to the first solution in the order given above. Agar is next added and dissolved at 100° C. The temperature is allowed to fall to 60° C. the mannitol is added, and the pH of the medium adjusted to 7.4 with N/10 NaOH. It is now "tubed" and sterilised at 15 pounds pressure for 15 minutes.

For the fungal counts a synthetic medium was also employed made up as follows:

K_2HPO_4	0.5 g.
Asparagine	1.0 g.
Glycerol	10.0 c.c.
Agar	15.0 g.
Water	1000 c.c.

The agar is dissolved at 100° C. and the pH adjusted to 4.8 with N/10 HCl. The medium is "tubed" and sterilised at 15 pounds pressure for 15 minutes.

With either Lemco or beef extract media it is impossible, owing to variations in the material, to obtain absolutely standard batches of cultures. On this account variations in bacterial numbers obtained on such media may well be due to variations in the composition of the medium. With a synthetic medium, such as that described above, it is possible to prepare each fresh batch of medium under completely standard conditions, and variations in bacterial or fungal numbers due to variations in the composition of the medium are eliminated. There is also the further fact that the presence of "spreaders," such as *Bacillus dendroides*, prevents accurate counts being made when a Lemco or beef extract nutrient medium is used. Their presence is very largely obviated when a synthetic mineral medium is employed.

For the fortnightly counts of both bacterial and fungal numbers, 10 plates of each medium (bacterial and fungal) were plated out and incubated at 25° C. for 7 days. In all cases χ^2 was calculated for each set of samples from the equation:

$$\chi^2 = \frac{S(x - \bar{x})^2}{\bar{x}},$$

where S stands for the summation over the whole sample, $\bar{x} = \frac{1}{n} S(x)$, n is the number of degrees of freedom and x is the mean of a number of observations. The conception of χ^2 is extremely important in connection with bacterial and fungal counts in estimating the numbers of these organisms by the dilution method, since it is possible by the use of this value to ascertain whether the distribution of the organisms follows the Poisson Series. An example of such a distribution as the Poisson Series is given by counts of yeast and bacterial cells in a culture where the distribution is necessarily discontinuous. It is possible by using the above equation and the tables given by Fisher (1934) to calculate significant deviations from expectation.

In neither of the two samples of flour (English and Manitoba) nor in any of the subsequent flours that were examined for bacterial

numbers, were the high figures recorded by Kent-Jones and Amos obtained. The highest figure recorded by us was 57,000 per gram.

The most important changes, biological and chemical, were found to occur, as would be expected, in the moistened samples. Here there was a rapid increase in soluble nitrogen (*i.e.*, soluble protein) and decrease in hydrogen-ion concentration, a fall in bacterial and an increase in fungal numbers. For example, the initial counts of bacteria at the commencement of storage (7.4.35) gave for the English low-grade sample at normal moisture content 40,750 per gram, and the corresponding value for the Manitoba sample was 37,800 per gram. Fungal counts were not taken at this time as a suitable technique had not been developed. Ten weeks later the bacterial numbers for the English normal sample were 14,400 per gram and the English moist sample 4,200 per gram, whereas the normal Manitoba sample gave 14,150 and the Manitoba moist sample 3,450 per gram. The fall in the bacterial numbers recorded for these samples corresponds with a decrease in the hydrogen-ion concentration over the same period of time, as may be seen from the figures given in Table I.

TABLE I

CHANGES IN pH AND BACTERIAL CONTENT OF FLOUR DURING STORAGE

English normal			English moist		
Period of storage in weeks	pH	Bacterial numbers per gram	Period of storage in weeks	pH	Bacterial numbers per gram
0	6.18	40,750	0	6.18	40,750
4	6.18	40,342	4	6.15	26,400
6	6.18	35,000	6	6.13	15,000
8	6.16	14,800	8	5.66	6,321
10	5.66	14,400	10	4.93	4,200
Manitoba normal			Manitoba moist		
0	6.24	37,800	0	6.24	37,800
4	6.27	37,600	4	5.93	14,800
6	6.21	25,500	6	5.17	4,500
8	6.19	14,856	8	5.14	3,900
10	6.14	14,150	10	5.33	3,450

It is a well known fact that bacteria do not multiply rapidly in a medium of less than pH = 6.0 and it is interesting to compare the rapid increase in hydrogen-ion concentration in the moist samples during storage with the especially great fall in bacterial numbers in the same samples.

From a large number of fungal counts on different flours, it has been found that the average initial values in freshly milled normal flours,

i.e., excluding very low-grade flours, lie between 1,000 and 2,000 per gram. In the moist sample of Manitoba flour, the fungal content rose from an initial value of 1,200 per gram to 8,280 per gram in 10 weeks, and the odour of mould was noticed after 8 weeks of storage. After 30 weeks of storage the fungal numbers had increased to 24,400 per gram and the bacterial numbers had fallen to 1,000 per gram. A marked increase in the fungal numbers was also observed in the moist English sample.

Over 90% of the fungi isolated from flours belong to the genus *Penicillium*, but other genera are also present, *e.g.*, *Botrytis*, *Aspergillus* and *Cladosporium*, as well as certain unidentified forms. The unidentified genera were examined at our request by S. F. Ashby, Director of the Imperial Mycological Institute, Kew, who suggested that they were very similar to certain forms that had been isolated from frozen meat by Brooks and Hansford (1922). Of the *Penicillia* present, *P. brevicompactum*, which forms mainly mycophenolic acid ($C_{17}H_{20}O_6$) from glucose in Roulin-Thom's medium, was fairly abundant, as well as a number of other species which also gave rise to mycophenolic acid, *e.g.*, *P. patris-mei* Zaleski. By far the greater number of *Penicillia* isolated from flour gave rise, however, on Roulin-Thom's medium to polymannose and polygalactose. A description of these products of fungal metabolism was first given by Clutterbuck *et al.* (1934) for a *Penicillium* isolated by them from mouldy maize, and to which they gave the specific name of *P. Charlesii*. This form is said by them to belong to the Monoverticillata-Ramigena group of Thom (1930). A number of *Penicillia* have been isolated by us from flour which give rise to these two polysaccharides, but which do not belong to this group. Some of these have been named for us. For example, one *Penicillium* (our laboratory number N.U. 17) was found to belong to Thom's Fasiculata-Viridicata group, while another (N.U. 18) was found to be identical with *Penicillium Expansum* (Thom's series) and yet another (N.U. 32) to Thom's *Aeruginosa* group and the nearest species with which it could be identified was *P. cyclopium* Westling. All these forms gave rise to polymannose and polyglucose when grown on Roulin-Thom's medium.

The fact that bacteria tend to disappear, and the numbers always fall to a very low value during prolonged storage of flour, whereas fungi increase rapidly in numbers if the moisture content of the flour be 16.0% or higher, explains why bacterial troubles are rarely encountered, if ever, during prolonged storage of flour. On the other hand, fustiness and mustiness due to mould growth are well known.

In the moistened samples of flour, the oil content (fraction soluble in petrol ether) was found to decrease rapidly during storage. The

values of the oil content in the moist samples of English and Manitoba flour are given in Table II. The initial iodine value for the oil from the English sample was 114.6 and for the Manitoba flour 112.4. Storage of flour did not significantly alter these values in either the normal or the moist samples.

In the next stage in the investigation the effect of storage on a low grade flour (ash content = 0.832%) was studied from a rather different point of view. This flour was stored at two different moisture contents (14.5 and 18.0%) and a portion of the moist sample was stored in presence of chloroform. It was found that the initial fungal content of this flour was abnormally high, 5,000 to 6,000 per gram as against a normal content of 1,000 to 2,000 per gram for higher grade flours. In the moist sample there was a rapid increase in fungal

TABLE II

CHANGES IN THE FAT CONTENT OF FLOUR DURING STORAGE

Period of storage in weeks	English flour (Moist)	Manitoba flour (Moist)
	% Oil content calculated on 15% moisture basis of original flour	% Oil content calculated on 15% moisture basis of original flour
0	0.910	1.340
2	0.870	1.220
4	0.880	0.940
6	0.728	0.660
8	0.455	0.534
10	0.197	0.430
12	0.100	0.330
14	0.085	0.228
16	0.065	0.153
18	0.066	0.098
20	0.064	0.074

numbers, the value being 200,000 per gram after 6 weeks and after 20 weeks of storage they reached the relatively enormous total of 1,250,000 per gram. The presence of chloroform in one of the samples inhibited the growth of all bacteria and fungi.

It should be mentioned here, that although in the various moistened samples of flour high fungal numbers were soon recorded, yet microscopical examination of the flours themselves showed no evidence of the normal mycelial growth of the fungi. Repeated examinations were also made for yeasts to see if they form a major part of the fungal content of moist flour. Up to the present, a fair number of yeast cells have been found, but the real explanation of the increase in fungal numbers would appear to be the fact that the spores of these forms, instead of germinating normally to give the usual thready myce-

lial growth, increase by "budding" in much the same way as yeasts. The starch grains in a flour heavily contaminated with fungi show these fungal spores firmly attached to the grains forming structures rather like necklaces.

Two unusual features were noticed with this low-grade flour. There was no increase in soluble nitrogen in any of the samples, normal, moist or chloroform-moist, and further, after 6 weeks of storage, the gluten of the normal and chloroform-moist sample broke up completely on washing. In the moist sample, however, the gluten steadily improved in quality for many weeks (see Fisher, Halton, and Carter, 1937). It was found that in the normal and chloroform moist samples, the oil content remained constant at 2.40% over 20 weeks of storage, whereas the oil content of the moist sample fell to 0.15% during this period.

It was concluded that the high oil content of the normal and chloroform samples was in some way influencing the quality of the gluten, and that the observed deterioration of the gluten was due to some chemical change occurring in the oil. This was substantiated by extracting the oil from the damaged sample with petrol ether, after which the gluten washed out satisfactorily. When the extracted oil, after the petrol ether had been distilled off, was added to a good grade London flour the gluten was severely damaged and broke up completely on washing.

It was considered that it was possibly the free fatty acids present in the flour oil that were adversely affecting the gluten, and the acid value of the oil was determined at fortnightly intervals in each sample immediately following upon the stage at which it was first noticed that the gluten was breaking up on washing out. The figures for the acid value for 10 weeks are given in Table III, together with the "specific acid value," *i.e.*, the percentage oil content × the acid value. It will be seen from these figures that the acid values of the oil extracted from

TABLE III

CHANGES IN ACID VALUES OF FLOUR FATS DURING FLOUR STORAGE

Period of storage in weeks	Normal sample		Chloroform-moist sample		Moist sample	
	Acid value	Specific acid value	Acid value	Specific acid value	Acid value	Specific acid value
2	52.1	124.6	47.6	104.4	63.2	17.72
4	54.2	129.4	49.1	107.1	57.6	11.86
6	56.1	135.0	51.7	113.4	64.0	8.91
8	58.4	146.1	53.9	117.5	48.2	7.34
10	64.6	155.3	59.7	131.7	66.1	7.84

the normal and chloroform samples increase steadily in value during storage, whereas the acid value of the moist sample tends to fluctuate, a fact possibly connected with the low total oil content, which made determination of the acid value difficult. Further, although the acid value of the oil from the moist sample is higher than that from either of the other two samples, the gluten did not break down on washing, but improved in quality with the age of the flour. Although the acid value of the moist sample was higher than either of the other two, the oil content of the flour had fallen from 2.4% to 0.28% when the first determination of the acid value was made. Presumably on this account, there was not sufficient concentration of free fatty acids present in the oil to damage the gluten. This was found to be the case. Oil extracted from the moist sample and added in 0.28% concentration to a good grade London flour did not damage the gluten. On the other hand, when the same oil was added in concentrations of 1.0 and 2.0% severe damage occurred and the gluten broke up on washing.

Since the acid value of the oil increases during the time of storage, it would appear that the oil is progressively hydrolysed into its free fatty acid constituents through lipase activity, and the lipase must come from the flour itself and not from either bacteria or fungi, since the chloroform sample proved to be completely sterile.

The question of whether it was the free fatty acids or the glycerides of flour oil which were responsible for the damage to gluten was further investigated. A large-scale extraction of the moist sample was made, using 11 pounds of flour which was extracted for 5 hours with petrol ether. The total yield of oil was 14.1 g. The extract was taken down under reduced pressure in an atmosphere of nitrogen and 8.5 g. of the crude product were dissolved in 100 c.c. of petrol ether; 50 c.c. of absolute alcohol were now added and the mixture was titrated against alcoholic potash. Fifty cubic centimeters of water were next added and the whole was well shaken in a separating funnel. The mixed petrol ether and water-alcohol fractions were separated off and both were well washed with successive lots of a mixture of 50% alcohol-water and petrol ether. The alcohol-water extract was acidified with dilute hydrochloric acid and the free fatty acids that separated out were taken up in petrol ether. The true glyceride was obtained from the evaporation of the petrol ether extract. In all, 3.1 g. of free fatty acids and 5.2 g. of glyceride were obtained. Both products were of a very dark brown colour. The fatty acid fraction remained liquid, but the glyceride fraction solidified and had a marked musty odour. Thus there were present in the oil of this sample 36.5% of free fatty acids and 61.2% of glyceride.

Twenty-four hundredths gram of each of the free fatty acid and

true glyceride fraction, *i.e.*, 10% of the original oil content of the flour, were added respectively to 10-g. samples of a good grade London flour to test their effect upon the gluten. It was found that the gluten was severely damaged by the free fatty acids and could not be washed out and broken up on the machine. The addition of glyceride, however, only produced slight shortening of the gluten. The glyceride was repurified by further extraction with alcoholic potash and again tested on the gluten of the London flour. The same result as before was obtained, the gluten merely showing a small amount of shortening such as is usually observed when pure fats are added in such quantities to flour.

A large-scale extraction (11 pounds) of the chloroform sample was also made and the petrol ether extract separated into true glyceride and free fatty acids by the method described above. In this case 63.63% of glyceride was found to 33.45% of free fatty acids. The effect of the glyceride and free fatty acids was tried upon the gluten of the same London flour as was used in the examination of the fatty extracts from the moist sample, and the same results as those recorded above were obtained. The free fatty acids damaged severely the gluten, whereas the glyceride fraction merely brought about shortening.

Moulds in their action upon the fat content of flour appear to remove entirely the solid, *i.e.*, the saturated fatty acids first. A quantitative separation into saturated and unsaturated fatty acids can be made from a mixture of these acids by first preparing the lead salts and subsequently treating these with alcohol. It has been shown by Twitchell (1921) that the use of alcohol is to be preferred in this connection to the older method of using ether. The lead salts of the unsaturated acids are soluble in alcohol, whereas those of the higher saturated fatty acids are insoluble. An examination of the free fatty acids extracted from the moist sample of flour was made under the standard conditions laid down by Twitchell when it was found that the lead salts were entirely soluble in alcohol and no precipitate of lead salts of solid acids was obtained.

In view of these various results, it was considered desirable to carry out a further investigation of the behaviour of another similar sample of low-grade flour under storage conditions. Another sample of low-grade flour (ash content 0.665%) was obtained from the same mill. The initial moisture content of this sample was 15.69% and the oil content 2.11%. The iodine value was 120.0, the initial acid value 16.33 and the specific acid value 34.49.

A portion of the sample was moistened to 18% moisture content, and a further portion of the normal sample was stored in the presence of chloroform. Samples were removed for routine analysis at fortnightly periods as before.

Figure 1 shows the changes in oil content in the three different samples (normal, moist and normal/chloroform) over the period of storage. It will be seen that in the moist sample the oil content began to fall (and the fungal numbers to increase) after six weeks of storage. During this time the mould content had risen from the initial value of 2,300 per gram to 20,660 per gram. After 22 weeks of storage the oil content fell to 0.15% and the fungal content rose to its maximum figure of 503,000 per gram. Thereafter the oil content remained approximately constant at the low value of 0.1% and the fungal numbers began to fall. During the following 10 weeks the (fortnightly) fungal

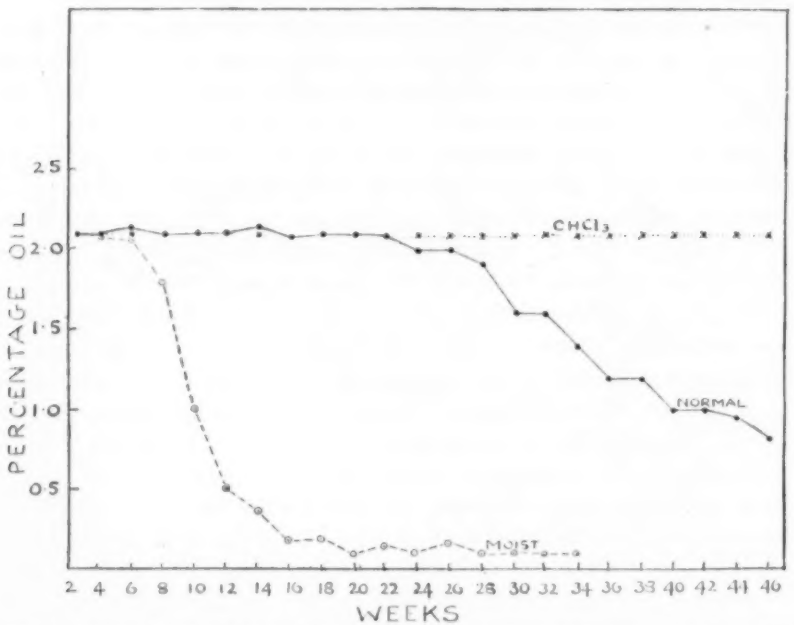

Figure 1. Changes in oil content of flour during storage: (*a*) at normal moisture content (15.69%); (*b*) at normal moisture content in presence of chloroform; and (*c*) at 18% moisture content.

numbers per gram fell and were as follows: 425,000; 381,000; 262,000; 122,000 and 94,000, the bacterial numbers increasing at the same time. During the 22 weeks when the fungal numbers were increasing, the hydrogen-ion concentration of the flour decreased in 12 weeks from its initial value of 6.33 to 4.12, and then increased to 6.27 in the succeeding 10 weeks. At the end of 22 weeks, therefore, the original hydrogen-ion concentration had been substantially regained (6.27), the fungal numbers had reached a maximum (503,000 per gram), and the bacterial numbers a minimum (800 per gram). Thereafter the hydrogen-ion concentration rose to 7.1 and the bacterial number rose to 620,000 per gram.

It is possible that the initial decrease in H-ion concentration is due to non-biological causes (*e.g.*, hydrolysis of phosphate), the subsequent increase being due to fungal activity.

The evidence suggests that the factor which determines whether fungi or bacteria shall grow in flour is the acidity: when the H-ion concentration is below a certain value only fungi will grow (bacteria dying off more or less rapidly); when the H-ion concentration is greater than this figure bacteria will flourish (while fungi will tend to disappear). Normal fresh flour has a H-ion concentration distinctly lower than is favourable for bacterial growth, which probably explains the observed decrease in bacterial numbers in most fresh flours during storage, a decrease probably accelerated by the decrease in H-ion concentration observed to take place in the earlier stages of storage.

In the normal sample, the oil content did not show any significant variation in amount until 22 weeks after storage. Thereafter, like the moist sample the value fell until after 54 weeks of storage it fell to 0.20%. This fall in the oil content is probably also due to increase in fungal numbers. The initial fungal content of the normal flour was 2,300 per gram and this rose in 20 weeks to 6,400 per gram. After 54 weeks the sample stored in the presence of chloroform shows no significant variation in oil content throughout the whole storage period (54 weeks).

Figure 2 shows the acid values for the oils of the three samples. It will be observed that in the moist sample the acid value rose to a maximum in 20 weeks and thereafter fell. In the normal sample there was a steady increase in the acid value up to 30 weeks of storage when the value began to fall away, while in the normal chloroform sample the acid value rose for the whole storage time. Figure 3 shows the specific acid values for the three samples. It will be seen that in the moist sample the specific acid value fell after six weeks of storage and finally reached a constant low value.

The behaviour of the gluten of this sample on washing out was not altogether the same as was experienced with the low-grade flour mentioned above. In the normal sample, after 4 weeks of storage the gluten showed a tendency to break up on washing, and this tendency increased at 8 weeks, but the gluten always came together again before washing was completed. At the same time the gluten was poor in quality, short, tough, and crumbly, and at the end of the storage period the gluten was very ragged, short, with no spring and would scarcely cohere; it decreased in amount from 12.7% to 7.48%. It is possible that the behaviour of the gluten here is due to mould activity and fall in oil content. On the other hand, in the normal/chloroform sample

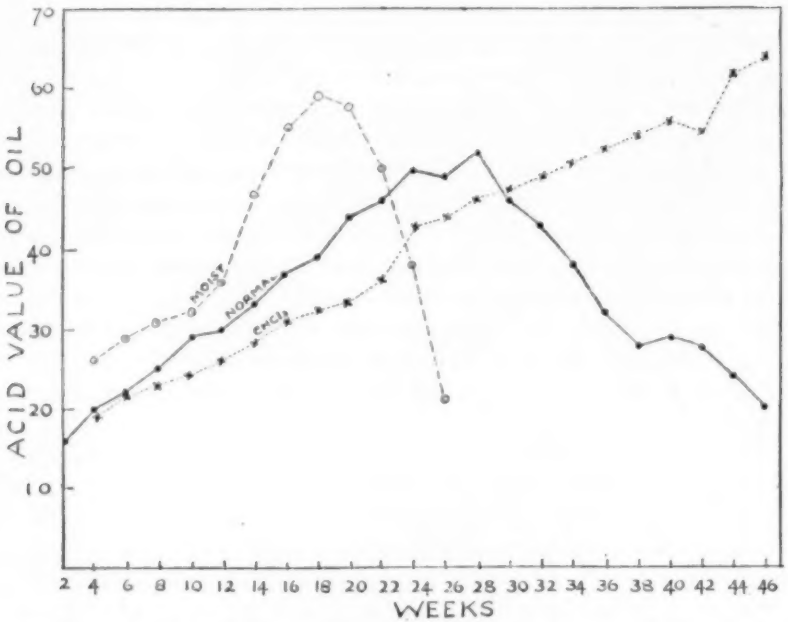

Figure 2. Changes in acid value of the oil in flour during storage: (*a*) at normal moisture content (15.69%); (*b*) at normal moisture content in presence of chloroform; and (*c*) at 18% moisture content.

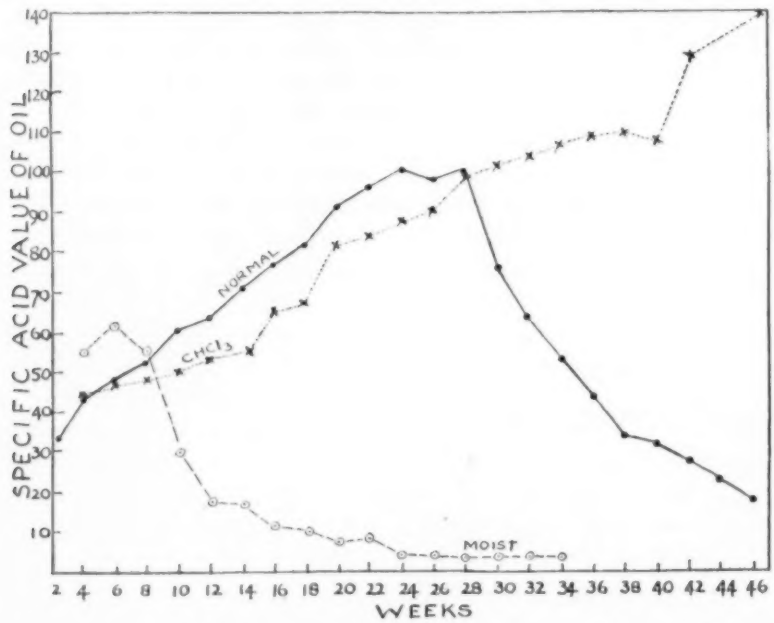

Figure 3. Changes in specific acid value (*i.e.*, acid value × percentage oil content) of the oil of flour during storage: (*a*) at normal moisture content (15.69%); (*b*) at normal moisture content in presence of chloroform; and (*c*) at 18% moisture content.

there was no alteration in the amount of oil during the whole period of storage and the acid value rose steadily during the same time, yet although the quality of the gluten was very poor, it never broke down completely in the same way as that from the former sample of low-grade flour.

Hartmann (1930) has claimed that it is possible to estimate the age of a flour by the apparently uniform increase in the acid value up to at least 15 months. The results obtained in this investigation show, however, that in normal flour on storage, the acid value increases to a maximum and then falls away, and that a continuous increase in the value only occurs in the presence of some sterilising agent like chloroform. Hartmann's claim cannot therefore be maintained.

According to Kosmin, Alkakvinokaja and Bondareff (1934), who stored different varieties of flours at different temperatues, such as 15° C., 30° C. and 45° C., the acid value of the oil content of the flours increases steadily, but at the lower temperature (15° C.) hydrolysis of the fat is much retarded, and there is no significant alteration in the iodine value. They also examined the effect of the oil of increased acid value on the condition of the gluten. Defatting of a flour which had been stored for two months with ether improved the quality of the gluten, which had previously shown signs of breaking up on washing. The gluten which prior to this treatment had crumbled away on washing was found to cohere to form a mass of elastic and silky condition. This agrees with the obserations made in this laboratory.

Since over 80% of the total fat content of flour consists of unsaturated fatty acids (*e.g.*, oleic, palmitoleic (?), linoleic and linolenic acids), these investigators tried the effect of pure samples of unsaturated acids, such as oleic acid on gluten. The acid was dissolved in ether and sprayed into the flour. The ether was allowed to evaporate off and the gluten was then washed out. The addition of oleic acid in amounts from 0.5 to 1.0% was found to have a marked influence upon gluten quality, the gluten becoming short, tough, crumbly and devoid of spring or extensibility.

It is clear that the addition of unsaturated fatty acids to flour has a remarkable effect upon the quality of the gluten, and the investigations to be described here have fully confirmed and extended the results obtained by Kosmin *et al.*

In the preliminary work upon the effect of unsaturated fatty acids on gluten quality, a pure sample of oleic acid was used and amounts between 0.1 to 2.0% were added to a good quality London flour. The oleic acid was dissolved in ether and sprayed into 10 g. of flour. The ether was allowed to evaporate off and the gluten was then washed out using a *rotor* gluten washing apparatus (Fisher and Halton, 1936).

It was found that concentrations of oleic acid up to 0.5% did not affect the quality of the gluten. With 0.5% there was a marked tendency for the gluten to break up on the machine, and the gluten was tough, short and had little extensibility. With 1.0% of oleic acid, the damage to the gluten was severe and it broke up badly on the machine and was only made to cohere with difficulty. With 2.0% of oleic acid the damage was even greater, and after washing the gluten remained in small lumps which scarcely cohered at all.

The question therefore arose as to whether the presence and number of double bonds in the molecule of the unsaturated fatty acids were the primary factors in affecting gluten quality. The unsaturated free fatty acid fraction of germ oil, for example, consists of 25 to 27% oleic acid, 50 to 55% of linoleic acid, and 3 to 5% of linolenic acid expressed as percentages of the total free acid. There is also present in the oleic acid fraction another unsaturated fatty acid with a single double bond in the molecule which appears to be the palmitoleic acid described by Lewcowitch (1914). The percentages of acids present in the mixture of unsaturated acids in various flour oils was estimated by means of the iodine and thiocyanogen values and the figures obtained would include such an acid in the oleic fraction.

Oleic acid has a single double bond in the molecule, linoleic acid two and linolenic three double bonds. A pure commercial sample of α-linoleic acid was obtained and varying amounts from 0.1 to 2.0% were added to a good grade London flour. It was found that even the lowest amount (0.1%) shortened and toughened the gluten, while severe damage was caused by 0.4% and the gluten broke up completely with the addition of 0.5%.

It thus appears that with an increase in the number of double bonds in the molecule of the fatty acid the damage to the gluten is increased. The matter was also tested in another way. A sample of germ oil was brominated in anhydrous ether in the ordinary way for the determination of the hexabromide value. The linolenic hexabromide which is insoluble in anhydrous ether was filtered off and dissolved in benzene and 0.5% of this substance was then added to the same London flour as was used in the other experiments. The addition of the hexabromide produced no effect upon the gluten. The linoleic tetrabomide was also obtained (fraction insoluble in petrol ether). This was first decolourised with Norite and recrystallised from ligroin. Varying amounts from 0.1 to 2.0% in ether were then added to the London flour. With the exception that the highest value (2.0%) produced a slight amount of shortening of the gluten, none of the severe damaging effects produced by α-linoleic acid itself was found. It would thus appear that it is the presence of the double bonds in the acids that in some way produces this damaging effect upon gluten.

The carboxyl group of the acid, however, also appears to play a part in the matter as is shown by the effect of sodium oleate on gluten quality. In this case no effect was produced even when the concentration was increased to 3.0%, although 0.5% of the acid itself has a marked damaging effect upon gluten. It thus appears that both one or more double bonds *and* a carboxyl group are necessary to produce the damaging effect upon gluten.

The possible effect of the length of the carbon chain in causing damage to gluten was also investigated. The possible influence of length of chain was determined by using saturated fatty acids of the acetic series. The results are shown in Table IV. A (+) sign indicates damage, (++) severe damage, and (−) no damage.

TABLE IV

INFLUENCE OF ADDITION OF SATURATED ACIDS, OF ACETIC ACID SERIES, TO FLOUR ON THE QUALITY OF THE WASHED OUT GLUTEN

Percentage Acid Added to Flour

Acid	Formula	0.1	0.2	0.3	0.4	0.5	1.0	2.0
Acetic	$C_2H_4O_2$	(−)	(+)	(+)	(+)	(++)	(++)	(++)
Propionic	$C_3H_6O_2$	(−)	(−)	(+)	(+)	(++)	(++)	(++)
N. Butyric	$C_4H_8O_2$	(−)	(−)	(−)	(+)	(+)	(++)	(++)
Isobutyric	$C_4H_8O_2$	(−)	(−)	(−)	(−)	(+)	(++)	(++)
Valerianic	$C_5H_{10}O_2$	(−)	(−)	(−)	(−)	(+)	(++)	(++)
Isovalerianic	$C_5H_{10}O_2$	(−)	(−)	(−)	(−)	(+)	(++)	(++)
Caproic	$C_6H_{12}O_2$	(−)	(−)	(−)	(−)	(+)	(++)	(++)
Caprylic	$C_8H_{16}O_2$	(−)	(−)	(−)	(−)	(−)	(+)	(+)
Lauric	$C_{12}H_{24}O_2$	(−)	(−)	(−)	(−)	(−)	(−)	(+)
Myristic	$C_{14}H_{28}O_2$	(−)	(−)	(−)	(−)	(−)	(−)	(+)
Palmitic	$C_{16}H_{32}O_2$	(−)	(−)	(−)	(−)	(−)	(−)	(−)
Stearic	$C_{18}H_{36}O_2$	(−)	(−)	(−)	(−)	(−)	(−)	(−)
Arachidic	$C_{20}H_{40}O_2$	(−)	(−)	(−)	(−)	(−)	(−)	(−)

It is clear from these results that the higher homologues, such as palmitic, stearic and arachidic acid have no effect upon gluten quality and that the damaging effect of the saturated acids decreases with increase in molecular weight.

It was observed that with the lower homologues the extent to which the gluten was damaged appeared to depend upon the length of time the dough was allowed to stand in water after the addition of acid. Thus with 0.5% of acetic acid added to the flour and 15 minutes standing of the dough in water, the gluten broke up during washing, but came together again later in the operation. When the dough was allowed to stand for 30 minutes in water, the gluten broke up very badly on washing and was only made to cohere with difficulty. After the dough had been allowed to stand for 45 minutes the gluten broke up completely.

The question as to whether the damage brought about by the lower homologues of the saturated fatty acids might not be due to pH changes in the dough was next examined. The pH of a dough made up with different percentages of saturated fatty acid, as well as in many cases the pH of the flour suspension, was examined. The values are given in Table V.

TABLE V

EFFECTS OF ADDITION OF CERTAIN SATURATED FATTY ACIDS IN THE pH OF DOUGHS

Acid	0.25% acid added. pH of dough	0.5% acid added. pH of dough
Control dough (no acid) pH: 5.83	—	—
Acetic	4.98	4.54
N. Butyric	5.13	4.73
Isobutyric	5.18	4.89
Valerianic	5.34	4.93
Caproic	5.35	5.19
Caprylic	5.61	5.43

The pH of the flour suspensions with the addition of some of higher saturated fatty acids was also determined. It was found that the addition of lauric or myristic acid in either 0.25 or 0.5% concentration to a flour suspension did not alter the pH significantly. With 1.0 and 2.0%, however, there was a slight decrease in the pH. The values are given in Table VI.

TABLE VI

EFFECT OF ADDITION OF CERTAIN SATURATED FATTY ACIDS IN THE pH OF FLOUR-WATER SUSPENSION

Acid	0.5% acid added. pH of suspension	1.0% acid added. pH of suspension	2.0% acid added. pH of suspension
Control suspension (No acid) pH = 65.1	—	—	—
Lauric	6.51	6.31	6.14
Myristic	6.51	6.42	6.27

The highest homologues, palmitic, stearic and arachidic, did not alter the pH of either dough or suspension over the control even in 2.0% concentration.

It will be observed that the acids higher than caprylic acid do not alter the pH of either dough or flour suspension materially except at a concentration of 2.0% in the case of lauric and myristic acids, and it is these higher homologues which have the least effect upon gluten quality. Lauric and myristic acids only affect gluten quality in a concentration of 2.0% and at this concentration they do produce a

slight fall in the pH of the flour suspension. The evidence, although not altogether conclusive, does suggest that damage to gluten when it occurs through the addition of saturated fatty acids is at least to some degree a pH phenomenon and is not of the same type as the damage brought about by the addition of unsaturated fatty acids. Further, it is unlikely that the length of the carbon chain as such is a factor in the problem. Thus, stearic acid, $C_{18}H_{36}O_2$, possesses the same number of carbon atoms in the molecule as oleic acid, $C_{18}H_{34}O_2$. Stearic acid does not affect gluten quality, whereas oleic acid in a concentration of 0.5% or higher causes severe damage. It has been suggested by Kosmin *et al.* (1934) that the effect of oleic acid on gluten is a pH one, but this seems extremely unlikely as the effect of oleic acid on dough pH is very slight. Thus, in one case it was found that the pH of a dough was 5.83, and the addition of 0.25% of oleic acid altered the value to 5.77 and with the addition of 1.0% of oleic acid the value fell to only 5.73.

It has been shown by Fisher and Halton (1933) that the addition of fat or oil to a dough resulted in an increased yield of gluten due to an increase of the non-protein content of the latter brought about by the retention of fat or oil by the gluten. Such fats and oils as lard, castor oil, arachis oil, olive oil, *etc.*, were used and added in 2.0% amounts in milk emulsions to the flour. In every case it was found that the increase in gluten, *i.e.*, in the percentage of washed out gluten, was due to absorption of fat, although in no case was the whole 2.0% of added fat or oil retained by the gluten. The amounts of added fat or oil retained by the gluten varied between 0.2 to 1.7%, *i.e.*, from 10 to 85% of the added fat was retained, the remainder being lost in the wash water. It was considered worth while to see to what extent the higher saturated and unsaturated fatty acids are retained by gluten. For this purpose stearic (saturated), oleic (unsaturated) and the mixed unsaturated fatty acids isolated from germ oil were added to a London flour and the washed out gluten analysed. The technique of oil extraction was the same as that used by Fisher and Halton. It is not possible to extract fats from gluten with either ether or petrol ether, so acetone was employed instead. The various fatty acids were added in 1.0% amounts, expressed by weight of flour, in ethereal solution. After the ether had been allowed to evaporate off, the gluten was washed out on the machine and extracted with acetone for fat estimation in the manner described by Fisher and Halton.

It was suggested to us that unsaturated fatty acids might be retained by gluten to a similar extent as lard, castor oil emulsion, *etc.*, are retained by it, and as a result of this increase in concentration during washing damage to the gluten may be intensified. This was

not found to be the case. The added fatty acids were found to be almost entirely washed out of the gluten. It is evident, therefore, that damage by unsaturated fatty acids to gluten is not primarily due to nor intensified by an increased concentration of these acids in the gluten itself.

Since unsaturated fatty acids, in concentrations of 0.5% and more, expressed as weight of flour, cause severe damage to gluten, the effect of adding these acids to doughs to test their effect on the baking quality of bread was next investigated. For this purpose the mixed saturated and unsaturated acids obtained from the hydrolysis of germ oil were used and sprayed into the flour in ethereal solution. In addition to the ordinary control (dough without addition of acids), the addition of 2% lard was also used, the lard being sprayed into the flour in ethereal solution in one case and "worked in" by hand kneading in the other. The baking tests were carried out with a low-grade flour and also with a good grade London flour. The results of the baking tests are summarised below:

Baking Tests with Low Grade Flour

(1) *Ordinary control.* The dough showed good body, extensibility and spring, and on baking gave a good all-round loaf.

(2) *Ether only added.* The ether was sprayed into the flour and then allowed to evaporate off overnight. The dough in this case also showed good body, extensibility and spring. The volume of the loaf was larger with crumb a trifle more open in grain than (1). The addition of ether alone gave a slightly improved loaf.

(3) *Lard* (2%). The dough showed good body and its spring and extensibility were excellent. The volume of the loaf was larger than either (1) or (2). The crumb was close and even in grain with good pile, texture and spring. Crumb colour was also improved over (1) and (2).

(4) *Saturated acids* (0.5%). The dough here was much shorter and tighter than (1), although receiving 1 gallon extra liquor per sack. The load showed a large "flying" top, but the volume was smaller than (1) and the crumb was "dead."

(5) *Saturated acids* (1.0%). The dough here was also short and tight, but the baking results on the whole were good. The load volume was excellent with very good face. Crumb showed good grain, texture and pile, but rather poor spring.

(6) *Unsaturated acids* (0.5%). The dough in this case as in (4) and (5) was short and tight. The volume of the loaf was poorer than (1) and the crumb was close and "dead."

(7) *Unsaturated acids* (1.0%). The dough was again short and tight. The volume of the loaf was slightly larger than (6), but crumb again was close and "dead."

(8) *Unsaturated acids* (2.0%). In this case also the dough was short and tight. The volume of the loaf, however, was excellent and considerably greater than (1), or (6) and (7). The crumb still showed the unsatisfactory characters obtained with the smaller additions of acid, *i.e.*, it was very close, "cheesy" and "dead."

Baking Tests with Good Grade London Flour

(1) *Control.* This gave an excellent all-round loaf.

(2) *Ether added.* The volume of the loaf, as was found in the case of the low grade flour, was slightly larger than the control (1) and the crumb was paler in colour.

(3) *Lard* (2.0%). The lard in this sample was "worked" into the flour. The volume of this loaf was the best in the set. The crumb possessed a close, even grain, with good texture and pile, and its colour showed an improvement on (1).

(4) *Lard* (2.0%). The lard in this sample was sprayed in ethereal solution and the ether allowed to evaporate off over night. The volume of this loaf was poorer than (3), but much improved on (1). The remaining characteristics were the same as in (3).

(5) *Saturated acids* (0.5%). The loaf volume was larger than (1), but the crumb was close, clay-like and "dead."

(6) *Saturated acids* (1.0%). The volume of the loaf here was greater than in either (1) or (5), but the crumb characteristics were the same as in (5), *i.e.*, close and even, but clay-like and "dead."

(7) *Unsaturated acids* (0.5%). The volume of this loaf and general appearance of the crumb were similar to (5).

(8) *Unsaturated acids* (1.0%). The volume of the loaf here was slightly greater than (7), but the crumb was again close, "cheesy" and "dead."

(9) *Unsaturated acids* (2.0%). The general quality of this loaf was the same as (8), but the colour of the crumb was poorer than the control (1), or (7) or (8).

It is clear from these baking tests that although the addition of unsaturated fatty acids does produce marked deterioration in such loaf qualities as volume, face and crumb, they do not damage a loaf to the extent that might be anticipated from their direct effect upon washed out gluten.

Summary

An investigation has been made of the changes that take place in the bacterial and fungal flora of flour kept under storage conditions. It was found that the bacterial numbers always diminish with increasing time. The fungal numbers, on the other hand, increased to a maximum when the moisture content of the flour was 16% or over. Deterioration leading to mustiness in flour with a high moisture content was found to be due to this increase in the fungal flora and not to the presence of bacteria.

Deterioration of gluten quality during storage of flour, especially low-grade flours, was found to depend on the unsaturated fatty acids present in the oil fraction. In flours with a moisture content of 16% and over, the oil content fell rapidly during storage and this was found to coincide with increase in fungal numbers and was not accompanied by deterioration in gluten quality.

In low-grade flours of normal moisture content (14 to 15%) or less with a high oil content it was found that after 6 to 8 weeks' storage the gluten tended to break up on washing, whereas in the moist samples, *i.e.*, with moisture content of 16% and over, the gluten improved in quality. This improvement in the quality of the gluten was found to be due to (1) the removal of fats and fatty acids by fungi and (2) some improving action due to the fungi themselves.

The effect of different saturated and unsaturated fatty acids on gluten is described and discussed. Saturated acids of high molecular weight produce only slight shortening effect on gluten. Unsaturated acids produce serious damage in gluten. This damage is due to the presence of double bonds in the acid since when these are removed, *e.g.*, by bromination, the damaging effects are removed.

Free carboxyl groups are also an essential factor in gluten damage since this is not caused by esters (*e.g.* fats themselves) or by salts of unsaturated fatty acids.

Gluten damage by unsaturated acids is not due to pH changes as the effect of, *e.g.*, oleic acid on dough pH is very small (0.1).

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A STUDY OF CERTAIN PHYSICAL PROPERTIES OF BREAD THROUGH THE STALING PROCESS WITH THE USE OF THE ELECTRONIC CELL

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Bakers Weekly, 45 West 45th Street, New York, New York

(Read at the Annual Meeting, May 1937)

A study of the literature reveals considerable data that relate to bread staling. Many of these data have to do with certain starch characteristics. There seems to be a relationship between the data available in the starch literature directly and the information that has been made available in the study of the staling of bread. Katz (1917) discussed the swelling and retrogradation of starch in connection with the staling of bread.

Samec (1928), in his studies of the physical and chemical characteristics of starch, has observed that the viscosity of the starch gel decreases with age. He attributes the cause of this alteration with aging to some colloidal variation in the dissolved substance which in aggregation and coagulation results in the precipitation of some substance that is responsible for the high viscosity. He also observed that the same degree of viscosity is not again obtained in restoring an aged starch gel. The assumption followed that the paste-forming type of starch substance (which has been previously designated by Maquenne as amylopectin) is irreversibly changed by aging. Samec assumed this amylopectin to be an electrolyte with a phosphorus complex. The

[1] The authors acknowledge the valuable assistance rendered by George Gaynor Hyde in cooperating in designing and constructing the apparatus.

aging process that caused a decrease in the gelatinous properties caused an increase in the electrical conductivity. His findings show that aging, heating under pressure, and demineralization all produced the same effect in changing a starch from a gelatinous form to one incapable of being gelatinized.

Nakomura (1935), in his work with starch gels, verified the results obtained by Samec.

Katz (1928), as well as Sherrer (1918) and Herzog and Jancke (1920), has shown that starch has a crystalline structure. Katz found a great similarity in the different varieties of starches, such as the starch of wheat, potatoes, rice and other grains. He offered the explanation that the same crystalline substances produced the same X-ray diagram but that differences were caused by admixtures or differences in the colloidal state of the substance. With substances of high molecular weight, such as cellulose and acetyl cellulose, while they vary in physical qualities, he obtained a similar or identical X-ray pattern. He felt, however, that in the case of starch definite conclusions as to its structure such as had been arrived at by those mentioned above should not be reached for two reasons: (1) because it is quite difficult with the type of diagram obtained from starch to be certain that in each sample the interferences completely coincide; (2) because there is still a possibility of the existence in the starch of various degrees of polymerization and that their X-ray diagrams show slight differences which heretofore have not been observed.

Katz is also in agreement with Samec's observation that a very intensely dried starch is more susceptible to the influence of hydrolysis than is a mildly air-dried starch. He states this follows from the fact that changes in the diagram of starch do not occur with the swelling or contracting but only when the substances are very thoroughly dried, in which case the diagram becomes definite and sometimes resembles the diagram of an amorphous phase.

Katz's theory is that there are two stages in the gelatinization of starch: first, when the starch reaches a temperature of 60 to 70° C. with an excess of water when a part of the starch becomes soluble, which causes an increase in the swelling; second, when a temperature of 100° C. is reached with a large amount of water present that will produce a starch paste. When kept at room temperature, or in a refrigerator without loss of water, the swelling power diminishes considerably in from one to two days. As this occurs there is a decrease in the soluble starch.

He states that when wheat starch is mixed with from 35 to 45% water and heated to 100° C., the ordinary X-ray pattern of the starch changes to a new one. He observes an interference in the X-ray

diagram through what he believes to be an effect of the ungelatinized starch portion. He states that ungelatinized starch changes into an X-ray pattern that appears to be less crystalline. This may be due to an amorphous state which can exist within the starch granule under the condition described.

Alsberg (1928), in his work with starch viscosity, found that wheat starch obtained from different types of wheat produced gels that differed in their viscosity. He corroborated Katz and others in their findings in the retrogradation value of starch gels and states that retrogradation is more rapid at low temperatures and that stale bread contains less soluble starch than fresh bread.

Since the work of some of these investigators indicates that there is a crystalline and amorphous state within the starch gel, and that reactions leading from one to the other may be reversible, or at least that there is a tendency to go into the more pronounced amorphous state during the retrogradation process, we assume that the opalescence of starch gel might also be changed. Microscopic and other examinations of gels over a period of time indicated that there is a tendency for the gel to become more opaque, or to go from the transparent to the opaque, during the aging period. To determine whether or not this is true the authors designed a photometer containing a photoelectric cell that might be used in ascertaining the quantity of light that would pass through starch gels under different conditions and in this way arrive at a means of measuring the density of the gel so far as light is concerned. The design of the photometer was developed jointly with George G. Hyde of the Hyde Laboratories, New York, N. Y.

Figure 1 shows the construction and character of the photometer. To the right we have a vertical beam of light that goes through a condenser, or lens, and then falls upon a mirror, throwing the light at right angles to its original path. From here it passes through a sliding device known as a carrier or sample holder, thence through a fixed polaroid disc up through a focusing lens used in photographic work. From this lens the light passes on through a filter holder designed for carrying various light filters and then through a polaroid disc calibrated from 0 to 90°, and from this point directly to the photronic cell. The polaroid discs are used as a diaphragm to standardize the quantity of light that is to fall upon the photocell.

Thus we have a means of measuring the quantity of light that will pass through a substance that might be inserted in the sample holder. On studying the light-transmission property of various starch gels the writers made up gels containing various quantities of starch and added from 0.1 to 0.2 of 1% toluene as a preservative. A measured quantity (10 c.c.) of stock gel was deposited into a special Petri dish measuring

7 cm. in diameter and 0.75 cm. in depth. To maintain a constant moisture content within the gel a glass cover designed for the purpose was used. This kept the gel within an enclosed chamber.

In determining the light penetration the photometer was first calibrated so that light of a known quantity was registered on the meter. The Petri dishes used in the investigation were also calibrated since it was found that there is a difference of from 18 to 20% in the penetration of light through dishes of this kind. After the proper calibration the Petri dishes containing the gel, minus the cover glass, were inserted in the holder shown at the left of the diagram. The

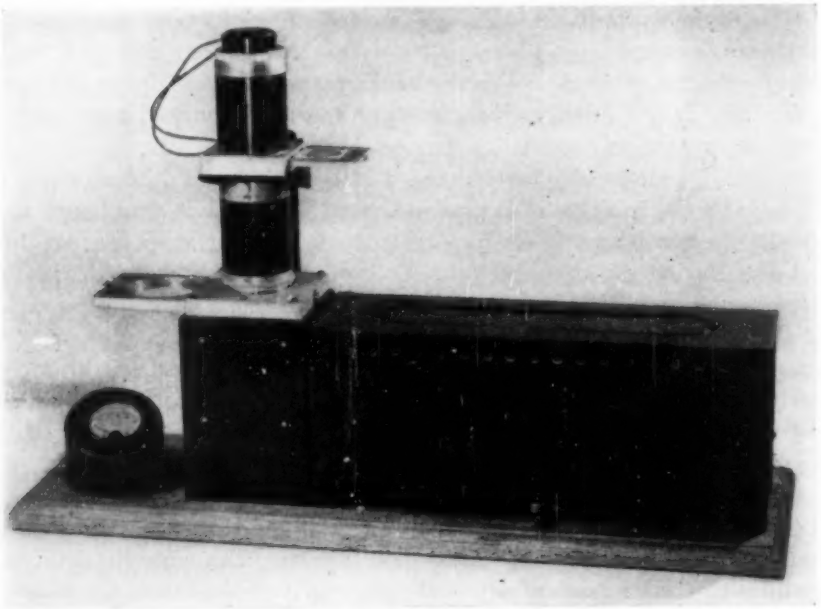

Figure 1

upper polaroid shown directly in the path of light in the diagram is used for calibration purposes. The light was then adjusted so that the dish containing the gel was in the direct path of the light beam, and readings were made with the gel in this state. The results obtained are given in Table I.

Table I shows that 5% and 10% gels were used. The readings were made three times daily for the first few days and twice daily thereafter over a period of ten days. It will be seen that records were maintained of temperature, humidity, and barometric pressure. Readings are given for wheat flour, wheat starch, corn starch, and potato starch. The quantity of light going through the sample is

recorded in microamperes or fractions thereof. It may be seen that the microampere readings are lowered as time goes on. In the wheat flour there is a decrease of 17.6%; in wheat starch, 32.3%; in corn starch, 30%; and in potato starch, 40.85%. Readings on the potato starch were terminated on the sixth day due to the starch gel becoming liquid at this point.

TABLE I

PHOTOMETER READINGS OF STARCH GELS[1]

	Temperature	Humidity	Barometric pressure	5% gels				10% gels			
				Flour	Wheat	Corn	Potato	Flour	Wheat	Corn	Potato
1st day P.M.	69.5	53	29.89	9.65	37.8	40.2	40.8				
2nd day A.M.	69.5	52.5	30.11	9.09	27.9	30.2	26.5				
2nd day M.	70.5	52	30.08	9.09	27.9	30.2	25.9				
2nd day P.M.	72.5	54.5	30.06	9.09	27.9	30.2	25.9				
3rd day A.M.	73.5	55.5	30.19	8.81	27.3	29.6	23 (Slight liquefaction)				
3rd day M.	74.5	54.5	30.15	8.81	27.3	29.6	22.7	8.5 Hot	14.3 Hot	32.2 Hot	17.1 Hot
3rd day P.M.	76	54	30.09	8.81	27.3	29.6	22.7	8.5	12.5	26.9	17.1
4th day A.M.	73.5	65.5	29.98	8.52	26.8	29.1	21.8	8.25	11.6	23.3	13.85
4th day P.M.	76.5	69	29.91	8.52	26.8	29.1	21.8	8.25	11.6	23.3	18.85
5th day A.M.	75.5	78	29.86	8.52	26.8	29.1	23 (Considerable liquefaction)	7.98	11.6	22.7	12.8
5th day P.M.	74.5	78	29.83	8.52	26.8	29.1	24.1	7.98	11.3	22.7	12.5
6th day A.M.	71	62	29.75	8.23	26.2	28.5	24.1 (Complete liquefaction)	7.98	11.0	22.4	11.7
6th day P.M.	71	56	29.82	8.23	26.2	28.5		7.98	11.0	22.4	11.45
7th day M.	71	53.5	29.94	7.95	26.2	28.5		7.98	10.7	22	11.18
8th day A.M.	69	57.5	29.85	7.95	26.2	28.5		7.98	10.7	22	10.62
8th day P.M.	71.5	57.5	29.83	7.95	26.2	28.5		7.7	10.7	21.7	10.62
9th day A.M.	71	56.5	29.98	molded	26.2	28.5		7.7	10.7	21.7	10.35
9th day P.M.	73	57.5	29.95		26.2	28.5		7.7	10.7	21.7	10.35
10th day A.M.	70.5	59.5	29.91		25.6	27.9		7.45	10.4	21.4	10.1 (Slight liquefaction)
Amount decreased				1.7	12.2	12.3	16.7	1.05	3.9	10.8	7.0
% decreased				17.6	32.3	30.6	40.85	12.37	27.25	33.55	40.90

[1] Readings in microamperes.

In the 10% gel the depreciation in the quantity of light that passed through was 12.37% for the wheat flour, 27.25% for wheat starch, 33.55% for corn starch, and 40.9% for potato starch. This shows a definite and appreciable reduction in the quantity of light that will

pass through starch gels of the character and density presented in the table.

From this we see that starch gels become more opaque as time goes on and that it becomes possible to measure the degree of opalescence with the photoelectric cell.

As early as 1852 Boussingault discovered that the staling of bread was not due to the drying out presented through the loss of moisture. He demonstrated by chemical analysis that bread was stale even though there was no appreciable loss of moisture. He placed bread in a water-saturated atmosphere so as to bring about the presence of water vapor, and under these conditions the bread became stale. He heated stale bread and brought it back to its state of freshness and then again repeated the staling process under the conditions mentioned. He states that by the reheating process the cycle can be repeated many times. In concluding his experiments he advanced the hypothesis that stale bread does not differ from fresh bread due to the loss of moisture but rather because a change in the molecular structure takes place and that this change begins when bread becomes cool.

Others since his time arrived at the same conclusion. Nothing definite was known of the underlying cause of staleness until the more recent investigations of Katz and others. Katz (1934), in a study of bread staling, determined the rate of staling by three methods; namely, (1) by testing the crumbliness and hardness with the fingers; (2) by changes in the swelling power presented in observations by Lindet (1902), Lehmann (1904) and others, through whose experiments it has been shown that crumb derived from stale bread swells less when placed in water than does fresh bread. In this method 10 g. of bread crumb, together with an excess of water, were passed through thin bolting cloth—usually 80 meshes per centimeter. The volume of liquid, which is saturated with toluene to maintain its sterile condition, is raised to 250 c.c. and the mixture allowed to settle in a graduated cylinder. At the end of 24 hours the volume of the sediment deposited is read off. After shaking again the sample is allowed to rest for another 24 hours for settling and the second reading is made. The mean of these two readings is then determined. The volume of sediment is shown to be appreciably larger for fresh bread than for stale bread.

As an example Katz gives 52 c.c. for fresh bread and 34 c.c. for stale bread. The authors corroborated this work of Katz by running the same experiment with their own bread and obtained a volume of 56 c.c. for fresh bread and 36.5 c.c. for stale bread.

(3) The quantity of soluble amylose that can be extracted from bread is also larger for fresh bread than for stale. In making the

soluble amylose determination the clear liquid that is obtained above the decantate in making the swelling determination is poured off, filtered until clear, evaporated to the minimum volume, and then precipitated with an excess quantity of alcohol. The precipitate is dried, with proper precaution, and then weighed.

Verschaffelt and van Teutem (1915) carried on an investigation to determine the cause for the crumbling property of bread not following directly the swelling and other characteristics of bread staling according to the results obtained by Katz in an earlier work (1911). They found that bread as much as 24 hours old showed very little difference from the crumbling of fresh bread but after the 24-hour period the crumbliness began to appear. They concluded that in fresh bread each starch granule is surrounded by a protein gel or a gluten gel, producing a sort of capsule that fits very closely around the starch granule, and that no soluble starch is found outside of the granule. In stale bread no precipitates are formed anywhere but the outlines of the granules become sharper because of the development of air tunnels, or channels, that occur between the starch granule and the gluten phase that surrounds them. They give measurements of the diameter of these channels.

Katz and Derksen (1933) show how the swelling and soluble amylose parallel the change of the X-ray pattern although no crumbliness of the cellular structure has as yet taken place. In the authors' work with light measurements it was found there was no change in the opalescence of the bread within 48 hours. The change began to take place in the third day's readings. This seems to parallel the results that Katz obtained in his X-ray work in comparison with the swelling and soluble amylose properties of bread.

To carry on the work with bread, standard formulas of two types were selected, one to produce water bread and the other milk bread. The formulas employed were:

	Water bread	Milk bread
	G.	*G.*
Flour	1,000	1,000
Water	660	660
Dry milk	—	60
Salt	20	20
Sugar	40	40
Malt	2.5	2.5
Shortening	40	40
Yeast	25	25

The bread was sliced at a designated time with a microtome type of slicer produced by the Hobart Manufacturing Company. It was possible with this slicer to obtain slices of accurate measurements. The

dimensions of the discs of bread employed were 6 cm. diameter by ½ cm. thickness. These slices were then placed in the Petri dishes described in the technic for determining the light penetration of starch gels and stored at room temperature, and under refrigeration at from 0 to −2° C. The results obtained in making these various determinations are given in Tables II, III and IV. Table II represents Series 1; Table III, Series 2; and Table IV, Series 3 of the bread investigations. In these tables, as in the previous work on starch gels, the temperature, humidity, and barometric pressure for each of the readings were recorded.

TABLE II

PHOTOMETER READINGS OF BREAD [1]

Series I

	Temperature	Humidity	Barometric pressure	Water bread room temperature	Water bread refrigerated at 0° C. Humidity, 70%	Milk bread room temperature	Milk bread refrigerated at 0° C. Humidity, 70%
1st day P.M.	69.5	53	29.89	60	50	70	62.5
2nd day A.M.	69.5	52.5	30.11	58.75	50	68.75	62.5
2nd day M.	70.5	52	30.8	58.75	50	68.75	62.5
2nd day P.M.	72.5	54.5	30.06	58.75	50	68.75	62.5
3rd day A.M.	73.5	55.5	30.19	55	50	68.75	62.5
3rd day M.	74.5	54.5	30.15	55	50	68.75	62.5
3rd day P.M.	76	54	30.09	53.75	48.75	68.75	62.5
4th day A.M.	73.5	65.5	29.98	52.5	48.75	67.5	62.5
4th day P.M.	76.5	69	29.91	52.5	48.75	67.5	62.5
5th day A.M.	75.5	78	29.86	50	47.5	66.25	61.25
5th day P.M.	74.5	78	29.83	50	47.5	65	61.25
6th day A.M.	71	62	29.75	48.75	45	63.75	60
6th day P.M.	71	56	29.82	47.5	43.75	62.5	60
7th day M.	71	53.5	29.94	43.75	43.75	60	60
8th day A.M.	69	57.5	29.85	42.5	42.5	58.75	58.75
8th day P.M.	71.5	57.5	29.83	41.25	42.5	57.5	57.5
9th day A.M.	71	56.5	29.98	40	41.25	56.25	57.5
9th day P.M.	73	57.5	29.95	40	41.25	56.25	57.5
10th day A.M.	70.5	59.5	29.91	40	41.25	55	57.5
Amount decreased				20	8.75	15	5.0
% decreased				33.33	17.5	21.45	8

[1] Readings in microamperes.

It will be noted that the light penetration changes appreciably over the period of the investigation. In Series 1 (Table II) we obtain depreciations of 33.33%, 17.5%, 21.45%, and 8%. The marked difference between samples 1 and 2 and samples 3 and 4 is due to the temperature at which the bread was stored, showing definitely that storage at the low temperature does retard opalescence in bread of this kind.

TABLE III

PHOTOMETER READINGS OF BREAD[1]

Series II

	Temperature	Humidity	Barometric pressure	Water bread, sponge dough, room temperature		Water bread, sponge dough, refrigerated at 0° C. Humidity, 70%		Water bread, straight dough, room temperature		Water bread, straight dough, refrigerated at 0° C. Humidity, 70%	
				1	1-A	2	2-A	3	3-A	4	4-A
1st day A.M.	73.5	55.5	30.19	60	52.5	50	58.75	53.75	45	47.5	50
1st day M.	74.5	54.5	30.15	60	52.5	50	58.75	53.75	45	47.5	50
1st day P.M.	76	54	30.09	60	52.5	50	58.75	53.75	45	47.5	50
2nd day A.M.	73.5	65.5	29.98	60	52.5	50	58.75	53.75	45	47.5	50
2nd day P.M.	76.5	69	29.91	60	52.5	50	58.75	53.75	45	47.5	50
3rd day A.M.	75.5	78	29.86	58.75	51.25	50	58.75	52.5	43.75	47.5	48.75
3rd day P.M.	74.5	78	29.83	58.75	50	50	58.75	51.25	42.5	47.5	48.75
4th day A.M.	71	62	29.75	57.5	48.75	48.75	57.5	48.75	40	45	47.5
4th day P.M.	71	56	29.82	55	46.25	48.75	57.5	48.25	38.75	45	47.5
5th day M.	71	53.5	29.94	52.5	43.75	47.5	56.25	43.75	36.25	43.75	46.25
6th day A.M.	69	57.5	29.85	50	41.25	47.5	55	41.25	35	42.5	45
6th day P.M.	71.5	57.5	29.83	48.75	40	47.5	55	40	35	41.25	43.75
7th day A.M.	71	56.5	29.98	47.5	38.75	46.25	55	37.5	33.75	41.25	43.75
7th day P.M.	73	57.5	29.95	46.25	38.75	46.25	55	37.5	33.75	41.25	43.75
Amount decreased				13.75	13.75	3.75	3.75	16.25	11.25	6.25	6.25
% decreased				22.90	26.15	7.5	6.38	30.25	25	13.16	12.50

[1] Readings in microamperes.

In the second series (Table III) we have bread stored at room temperature and at below 0° C. Here results were obtained that parallel those given in the first series.

In the third series (Table IV), where milk bread is under investigation under the conditions previously stated, results again were obtained that parallel those of the previous investigation although it will be seen that the milk bread changes to a lesser degree than does water bread in this particular property.

From this investigation it may be concluded that there is a change in the property of opalescence in starch gels and in bread of the type

TABLE IV

PHOTOMETER READINGS OF BREAD[1]

Series III

	Temperature	Humidity	Barometric pressure	Milk bread, sponge dough, room temperature		Milk bread, sponge dough, refrigerated at 0° C. Humidity, 70%		Milk bread, straight dough, room temperature		Milk bread, straight dough, refrigerated at 0° C. Humidity, 70%	
				1	1-A	2	2-A	3	3-A	4	4-A
1st day A.M.	73.5	55.5	30.19	50	55	60	50	50	60	50	47.5
1st day M.	74.5	54.5	30.15	50	55	60	50	50	60	50	47.5
1st day P.M.	76	54	30.09	50	55	60	50	50	60	50	47.5
2nd day A.M.	73.5	65.5	29.98	50	55	60	50	50	60	50	47.5
2nd day P.M.	76.5	69	29.91	50	55	60	50	50	60	50	47.5
3rd day A.M.	75.5	78	29.86	50	55	60	50	50	60	50	47.5
3rd day P.M.	74.5	78	29.83	48.75	53.75	60	50	48.75	58.75	50	47.5
4th day A.M.	71	62	29.75	47.5	51.25	58.75	48.75	45	57.5	48.75	46.25
4th day P.M.	71	56	29.82	46.25	50	58.75	48.75	43.75	65.25	48.75	46.25
5th day M.	71	53.5	29.94	43.75	48.75	58.75	48.75	42.5	53.75	48.75	46.25
6th day A.M.	69	57.5	29.85	43.75	48.75	58.75	48.75	41.25	53.75	48.75	46.25
6th day P.M.	71.5	57.5	29.83	42.5	47.5	58.75	47.5	40	53.75	48.75	45
7th day A.M.	71	56.5	29.98	41.25	46.25	57.5	47.5	38.75	52.5	47.5	45
7th day P.M.	73	57.5	29.95	40	46.25	57.5	47.5	38.75	51.25	47.5	45
Amount decreased				10	8.75	2.5	2.5	11.25	8.75	2.5	2.5
% decreased				20	15.90	4.17	5.0	22.55	14.55	5.0	5.2

[1] Readings in microamperes.

investigated; that the change takes place gradually over a period of from 1 to 10 days; that it is possible to devise a photometer that can be used for measuring this particular property; that the changes in the property investigated parallel the change in crumbliness and the change that takes place in soluble amylose investigated by Katz and Katz and others; that further investigation may be carried on to determine the cause for the change in the opalescence of starch gels and in bread systems.

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DAVID AUGUSTUS COLEMAN
1892–1938

(Read at the Annual Meeting, Cincinnati, Ohio, May 1938)

David A. Coleman, Editor of *Cereal Chemistry*, member of the United States Department of Agriculture, died on February 25, 1938. Most of us know him as the highly successful editor of our journal, Chairman of our Malt Committee, a scientist of standing and an efficient member of the United States Department of Agriculture. But Dr. Coleman was much more than these. We would all be interested to know more about him and his work. We all owe him more than we perhaps realize.

What did he stand for in the United States Department of Agriculture? What services did he render to the Association of Cereal Chemists, and in what way did he advance the science of cereal chemistry?

David Coleman was born in 1892 on a farm at Natick, Massachusetts. He was reared in an agricultural environment which was reflected in his whole subsequent career. He entered the Massachusetts Agricultural College in 1910, graduating in 1914 with major credit in agricultural chemistry. His education was continued at Rutgers University, where he received his Ph.D. in 1917. His research dealt with soil bacteriology.

He joined the United States Department of Agriculture in 1917 where he remained until his death, with the exception of one year in the Medical Corps of the United States Army during the War. In the Department he advanced from one position to another until he became head of the milling, baking and chemical laboratory of the Grain Division of the Bureau of Agricultural Economics.

His colleague, E. C. Parker, has prepared an excellent memorial to Dr. Coleman from which I have drawn freely. Parker explains the central theme of Coleman's work in the Department. He says in this work Coleman "soon displayed an exceptional ability to interpret the significance of fundamental chemistry and physics research in conjunction with the practical commercial problems of grain handling, storage, processing and market evaluation, and to make a practical application of scientific truths to grain-inspection and processing problems." His activities were many, and numerous bulletins ap-

peared with Coleman as author or co-author. However, they were all characterized by an application of already discovered scientific facts to practical problems. In this he performed a great service.

The bulletins bearing his name suggest also his principal fields of work. Many of these bulletins have become the standards in their fields, helping those entrusted with practical work to obtain the benefits of scientific methods. Some of these are:

In 1926, with E. G. Boerner, "The Brown-Duvel Moisture Tester and How to Operate It."

In 1926, with Fellows and Dixon, "Testing Wheat for Protein, with a Recommended Method for Making the Test." Parker says: "This fine and useful research work and publication brought to Dr. Coleman the first national recognition of his unusual ability to make practical application of science in the field of market evaluation of grain."

In 1930, with several associates, "Milling and Baking Qualities of World Wheats."

In 1937, with Fellows and Zeleny, "Rapid Determination of Oil Content and Oil Quality in Flaxseed." This bulletin placed these scientific determinations within reach of the industry.

Dr. Coleman devoted a great deal of his time to the study of problems connected with the malting of barley. He installed at the Bureau's laboratories a most efficient small-scale experimental malting plant from which were discovered many important facts regarding the malting process. Here again he succeeded in making practical application of scientific research in the field of the market evaluation of grain.

One of the most useful achievements in the application of science to practical grain testing was Coleman's work on the determination of moisture in grain by the application of the principle of electrical resistance. Working in cooperation with the manufacturers of the Heppenstall moisture meter, he and his associates perfected the now well-known Tag-Heppenstall meter, worked out its adaptation to the determination of moisture in grain, and secured its widespread adoption.

In all this Coleman rendered important service to Federal Grain Supervision in the administration of the United States Grain Standards Act.

The success of associations like that of the Cereal Chemists depends upon whether or not they can enlist the active interest of able men. Dr. Coleman was one of the abler members of the A. A. C. C. who contributed greatly to its success. The sum of his contributions makes an imposing total. Coleman was a member of the Committee on Standardization of Laboratory Baking for 4 years, being Chairman in 1931. He was Chairman of the Committee on Methods of Analysis in

1926. He has been Chairman of the Malt Analysis Standardization Committee since its inception in 1934. His good judgment assisted the committee to avoid the pitfalls which surrounded it and to achieve useful results. This year Coleman was also a member of our Convention Program Committee. He added strength to each committee of which he was a member and made many helpful contributions to its work.

It is, however, as Editor-in-Chief of our journal that Dr. Coleman probably performed his principal service to the science of cereal chemistry and to our Association. When he took over the editorship from Dr. Bailey in 1931 the journal was already firmly established and in a thriving condition. Under his editorship it continued to thrive, to expand and to maintain itself as the leading journal of cereal chemistry in the world. Under his direction the size of the journal increased 50%, and the circulation in a similar manner. During this period he maintained the high scientific standards of his predecessor, and attracted the leading articles on this subject from North America and a number from foreign countries.

The editorship of a journal of this kind presents many difficulties. Dr. Coleman conducted its business efficiently and economically. While he properly did everything in his power to advance the interests of *Cereal Chemistry*, he was always fair and reasonable in his dealings with the other activities of our Association.

The officers of the A. A. C. C. always regarded the progress of the journal as assured so long as it remained in the hands of Dr. Coleman. In all of this work he freely acknowledged his debt to his associates.

David Coleman was everything that a man in his position should be. As a scientist he was well informed, progressive and scrupulously honest. In the Department of Agriculture he was highly regarded. As a friend he was always helpful and generous. In Dr. Coleman's death we have all suffered a great loss.—WASHINGTON PLATT.

PRESIDENTIAL MESSAGE

C. H. Bailey

University of Minnesota, St. Paul, Minnesota

(Read at the Annual Meeting, Cincinnati, Ohio, May 23, 1938)

There seem to be two principal reasons for a message from your President on the occasion of the annual meetings of this Association. The first involves an accounting of the year's activities, since the President is in a favorable position to survey these during his term of office. The second is to endeavor to make suggestions for the future activities of the organization as these are indicated by the experiences of the year just closed. Many of these matters are covered in the report of the Executive Committee, however, and need not be repeated here.

In assuming this office a year ago it occurred to me that our Association is somewhat like a great ocean liner in midsea. The ship is already manned with a trained crew, competent to discharge all the usual tasks. A new navigating officer in the person of the President comes onto the bridge to serve a brief tour of duty. Since the course has already been laid by his predecessors in office, the ship might sail on this course whether or not the officer gave strict attention to his duties. However, if truly alert to his responsibilities he insures that all the crew are at their posts and endeavors to distribute fairly the various tasks to competent persons. Should real emergencies arise he must direct the way out. It is conceivable that he might wreck the ship, but this is so improbable as scarcely to constitute a real hazard, particularly since all his acts are under the scrutiny of others fully competent to take his place.

To continue this analogy, your retiring President has encountered no serious storms during the past twelve months. Everyone who has been asked to perform a duty has done so cheerfully and well. When I step from the navigating bridge, and turn the task over to my successor, it will be with a feeling of real gratitude to all those who have labored hard and effectively for our professional advancement.

It is inevitable that some problems will arise over a span of twelve months, however, and those to whom the direction of the ship is charged can not shrink from making necessary decisions and discharging certain duties. New ideas, like breezes at sea, flow in upon us and we must reset our helm to suit the varying pressures.

A year ago Mr. Liggitt in his message emphasized the desirability of effecting a greater continuity in certain Associational activities. Since these tasks are largely handled through committees, this reduces itself to a plan whereby certain members are encouraged to remain on committees for a fairly extended period of time. During the current period an effort has been made to promote this program, but obviously

it will mature only through the sustained support of later administrations and of the members involved. While this may impose heavier tasks on these committeemen than heretofore, it will have the effect of developing a group of specialists who understand the technical problems thoroughly and can guide the Association in its formulation of methods and policies. When a member, for any reason, must discontinue his committee work, he should transmit to his successor fairly definite suggestions respecting incompleted tasks, that the program may go forward with greater definiteness and continuity. My recommendation is that Mr. Liggitt's proposal be developed further in the year just ahead, that we may add to our experience in this new practice.

Any administration as it nears its close doubtless endeavors to appraise its accomplishments and particularly as these involve anything new or distinctive. If the past year has registered an expansion of former activities, this may be found in the establishment of increasingly closer ties with foreign groups and institutions. Your President has been singularly fortunate in having the opportunity to attend in recent months international meetings in Leipzig, Germany; Scheveningen, Holland; and Oslo, Norway. Thus he was enabled to bring the greetings of our Association to the European cereal chemists and to participate actively in discussions involving formulation of methods and techniques and the interpretation of results.

The Vice-President has also been in close touch with the European, and more definitely the English groups on at least two occasions recently, and has visited numerous foreign institutes and laboratories. From these contacts has grown an enhanced interest in and appreciation of the views held in remote centers of cereal chemistry research and industry. Possibly these contacts will strengthen our own local work, and can be developed into a more general program involving many lands. It does appear that our Association has gained the respect of foreign workers. Now the increasing activities of certain European groups constitute a challenge to us to the extent that we in America must look to our laurels and insure that our future work is maintained on a scientifically sound and scholarly plane, else we may well be surpassed by our colleagues across the sea. This is a healthy situation, however, as is true of all honest competition, and should have the effect of stimulating us to our best efforts.

Now we are assembled for another national meeting, which, thanks to the local and other groups that have made the various arrangements, gives every promise of meeting the high standards set on former occasions. These general meetings mean much to the sustained life of such an association.

It is the steady driving forward which advances a profession and identifies its members as alert and useful scientists and technicians. The programs which this Association provides at its annual and other meetings are most serviceable in promoting this steady advance.

MINUTES OF THE TWENTY-FOURTH ANNUAL MEETING OF THE AMERICAN ASSOCIATION OF CEREAL CHEMISTS

Netherland Plaza Hotel, Cincinnati, Ohio

May 23 to 27, 1938

J. M. Doty, *Secretary*

An excellent attendance and a great enthusiasm imbued the opening of the Twenty-Fourth Annual Meeting of the American Association of Cereal Chemists in the Netherland Plaza Hotel, Cincinnati, Ohio.

President C. H. Bailey called the meeting to order at 9:30 a.m., Monday, May 23, 1938. The opening invocation was given by the Reverend L. W. Almy.

Washington Platt read a memorial to D. A. Coleman calling to mind his loyal and unselfish service to the Association during his many years of active membership. The names of other members who had died during the past year were read. These were E. E. Werner, J. R. Katz, H. W. Hahn, O. B. Winter, and Charles J. Henry. All members stood in silence a moment in their memory.

Dr. Bailey then introduced the Hon. James G. Stewart, Mayor of Cincinnati, who welcomed us all to Cincinnati, the Queen City.

A telegram from C. W. Partridge, Secretary of the Association of Operative Millers, extending good wishes for a successful meeting, was read. Leslie Olsen sent regrets that he was unable to attend and extended his wishes for a most successful meeting. Rowland J. Clark expressed his regrets that he could not meet with us and wished us a successful and inspiring meeting. C. C. Fifield sent a message explaining that the serious illness of his father would prevent his attendance.

W. H. Cathcart, representative of the American Institute of Baking, and Roger Brandenburg, President of the American Society of Bakery Engineers, were then introduced.

Dr. Bailey then read his Presidential Message, printed on pages 555–556 of this issue of CEREAL CHEMISTRY.

Dr. Bailey then introduced H. G. Knight, of the United States Bureau of Chemistry and Soils, who presented the paper "Research in Cereal Industry."

Our guest speaker from the American Society of Bakery Engineers, L. E. Caster, was then introduced by Dr. Bailey and presented a paper, "How Can Cereal Chemists Best Co-operate and Be of Service to Production Men." Mr. Caster brought home to us in convincing manner the importance of closer co-operation with and more actual experience in the bakeshop.

Paper—"Quantitative Methods for Evaluating the Quality of Macaroni Products," by D. S. Binnington and W. F. Geddes, read by Mr. Binnington.

Paper—"Some Observations Regarding the Flavor of Bread," by J. C. Baker and M. D. Mize, read by Dr. Baker. Discussion by Dr. Read.

Paper—"The Cumulative Development of Bakery Taste," by J. A. Dunn. Question by Mr. Mitchell.

Paper—"Temperatures Attained in Baking Products," by R. A. Barackman. Discussion by R. C. Sherwood and J. C. Baker.

At this time H. K. Loving made an announcement about the plans for the picnic for the afternoon.

Paper—"Recent Research on the Problems of the Baking Value of Rye Flours by European Cereal Chemists," by C. W. Brabender. Question by W. L. Green.

Mr. Reiman, Chairman of the Local Arrangements Committee, made a few special announcements. Dr. Bailey then recessed the meeting at 12:38 p.m. until 8:30 the next morning.

Tuesday, May 24

President Bailey reopened the meeting at 8:55 a.m. and introduced the presiding chairman, Elsie Singruen, and co-chairman, H. W. Rohde, in charge of the Malting and Brewing Session in the Hall of Mirrors. Running concurrently with this session in Parlor G was the Soft Wheat Symposium presided over by E. G. Bayfield.

Miss Singruen expressed her appreciation for the cooperation of the members of the Malting Analysis Standardization Committee. She then read the report of that Committee. Discussion by A. D. Dickson.

H. W. Rohde introduced the rest of the speakers on this program.

Paper—"A Comparison of Methods for the Determination of Diastatic Power in Malts," by A. D. Dickson and G. M. Burkert, read by the latter. Discussion by Mr. Austin, C. N. Frey, S. Laufer, and Elsie Singruen.

Miss Singruen called on Mr. Sippel, a member of the Master Brewers Association. She then introduced Marcus Maegerlein, past president of the Master Brewers and at present the Chairman of the Materials Improvement Committee of that organization.

Paper—"Present Day Problems," by Marcus Maegerlein.

Paper—"An Ultra-Centrifugal Analysis of the Degree of Lintner Soluble Starch," by Quick Landis and C. O. Beckman, read by Dr. Landis.

Paper—"I. Varietal Differences in Barleys and Malts. II. Diastatic Activity of Barleys," by H. R. Sallans and J. A. Anderson, read by Dr. Sallans. Question by Dr. Laufer.

Paper—"Barley and Malt Studies. V. Experimental Malting of Barleys Grown in 1937," by J. G. Dickson, H. L. Shands, A. D. Dickson, and B. A. Burkhart. Dr. Shands read the paper. Discussion by S. Laufer and M. Maegerlein.

Paper—"Determination of the Proteolytic Power of Malt," by L. E. Ehrnst. Question by Dr. Ford.

Paper—"Studies in the Mashing Process. I. Degradation of the Proteins During Mashing," by S. Laufer.

Paper—"Cereals Used in Brewing," by Elsie Singruen.

At this time the group from the Soft Wheat Symposium joined the Malting and Brewing Session.

Paper—"The Cereal Amylases, with Reference to Flour and Malt Behavior," by M. J. Blish, R. M. Sandstedt, and E. Kneen. Dr. Blish read the paper. Discussion by Dr. Malloch and Mr. Richards.

Paper—"A Simple Method for the Determination of Fermentable Starch in Grain," by M. C. Markley and W. B. Harr. Question by Dr. Laufer.

Miss Singruen then introduced a guest speaker, G. S. Sperti.

Paper—"Biodynes—Metabolic Stimulating Factors," by G. S. Sperti, J. R. Loofbourow, and E. S. Cook, each of whom presented information about the subject in a very interesting manner.

The Soft Wheat Symposium opened at 8:40 a.m. in Parlor G and Dr. Bayfield, presiding chairman, started the program by introducing B. B. Bayles.

Paper—"Getting the Right Variety in the Right Place," by B. B. Bayles.

Paper—"The Technique of Producing New Wheat," by W. W. Worzella.

Paper—"Environment and Wheat Quality," by E. G. Bayfield.

Paper—"Soft Wheat Milling," by R. Sopher. General discussion.

Paper—"Laboratory Testing Problems," by G. L. Alexander. Discussion.

Paper—"Utilization of Types of Soft Wheat," by H. M. Simmons.

Dr. Bayfield closed the Soft Wheat Symposium at 11:15 a.m. so those present might attend the Malting and Brewing Session and hear the last papers presented there.

The general session was recessed at 1 p.m.

Tuesday afternoon was taken up with inspection trips, and the golf and pistol tournaments.

(Tuesday's program for the ladies included a sightseeing trip at 10 a.m.)

Wednesday, May 25

President Bailey called the meeting to order at 8:43 a.m. and asked for the minutes of the Twenty-Third Annual Meeting of the Association. Since these had been printed in CEREAL CHEMISTRY, **14**: 587–603 (1937), they were referred to but

not read, and were approved without objection or correction. R. C. Sherwood moved that they be approved; seconded by Washington Platt; carried.

In the absence of D. A. MacTavish, Chairman of the Membership Committee, its report was read by Bert Ingels who moved its acceptance; seconded by J. A. Dunn; carried. This report may be found on page 567.

Dr. Geddes gave the report of the Executive Committee which appears on page 563. Dr. Geddes moved acceptance; Bert Ingels seconded; carried.

Paul Logue then read the report of the Committee on the Osborne Medal Award shown on page 566. J. A. Dunn moved acceptance; E. B. Brown seconded; carried.

The report of the Inter-Relations Committee, found on page 565, was read by R. C. Sherwood and he moved acceptance; seconded by W. L. Heald; carried.

H. K. Parker reported the work of the two Auditing Committees and moved it be accepted; seconded by R. C. Sherwood; carried.

R. W. Mitchell reported the work of the History Committee, shown on page 564, and moved it be accepted; seconded by R. C. Sherwood; carried.

The report of the Requirements for Membership Committee was presented by R. C. Sherwood. This report appears on page 565 of this issue. Dr. Sherwood moved that all those in sympathy with this report be asked to express themselves; seconded by H. D. Liggitt, Jr. All those present except two indicated their approval of the findings of this Committee. These findings of the Requirements for Membership Committee will necessitate a change in the Constitution in the form of an amendment.

Paul Logue reported the work of the Investment Committee, page 566, and moved that his report be accepted; seconded by W. L. Heald; carried.

The report of the Publicity Committee, page 567, was read by the Chairman, Victor E. Marx, and he moved that the report be accepted; seconded by W. R. Green; carried.

C. G. Harrel read the report of the Nominating Committee and moved the report be accepted; seconded by R. C. Sherwood; carried. Their recommendations and nominations follow: for President, W. F. Geddes; for Vice-President, G. F. Garnatz, W. L. Heald, and C. N. Frey; for Secretary, J. M. Doty; for Treasurer, Oscar Skovholt; for Editor-in-Chief of CEREAL CHEMISTRY, M. J. Blish; and for Managing Editor of CEREAL CHEMISTRY, R. M. Sandstedt.

Elise Shover moved that the nominations for President be closed and that the Secretary be instructed to cast a unanimous vote for W. F. Geddes; seconded by W. R. Green; carried.

M. A. Gray moved the nominations for Vice-President be closed; seconded by J. A. Dunn; carried. The balloting resulted in the election of G. F. Garnatz as Vice-President of the Association.

Nominations were open for Secretary and H. W. Putnam moved the nominations be closed and a unanimous vote be cast; seconded by H. C. Gore; carried.

M. D. Mize moved the nominations for Treasurer be closed and a unanimous vote be cast for Oscar Skovholt; seconded by R. W. Mitchell; carried.

Unanimous ballots were cast for M. J. Blish and R. M. Sandstedt on actions moved by R. C. Sherwood and D. S. Binnington, respectively, and seconded by M. A. Gray and M. D. Mize.

C. A. Glabau then read the report of the Employment Committee and moved the report be accepted; seconded by Elise Shover; carried.

Oscar Skovholt wanted to go on record that a new and revised edition of *Cereal Laboratory Methods* must soon get under way. He suggested that some action on this matter be taken very soon.

G. F. Garnatz, the newly elected Vice-President, then addressed the members and thanked them for the honor given him and promised to do his best to serve the Association.

C. H. Bailey then turned the meeting over to R. T. Bohn, presiding chairman, who presented the following speakers:

Paper—"General Report of the Committee on Standardization of Laboratory Baking," read by C. F. Davis, Chairman. Dr. Skovholt moved acceptance of the report; seconded by Dr. Malloch; carried.

Paper—"The Question of Sugar Levels in Laboratory Baking," by R. M. Sandstedt and M. J. Blish. Mr. Sandstedt read the paper. Questioned by Drs. Malloch, Moen, and Geddes.

Paper—"A Study of the Suitability of Sponge Procedures for Test Baking,"

by J. Freilich and Quick Landis. Mr. Freilich read the paper. Discussion by C. H. Bailey, Oscar Skovholt, and R. M. Sandstedt.

Paper—"Hand Punching and Hand Moulding *versus* Machine Punching and Machine Moulding," by W. L. Heald. Question by Dr. Malloch.

Paper—"A Cooperative Test of a Punching and Moulding Machine," by J. G. Malloch. Discussion by W. L. Green.

Paper—"A Practical Method for Utilizing the Tentative A. A. C. C. Baking Test," by M. C. Markley.

Paper—"A Critical Study of a 'Pup' Sponge Baking Method," by J. A. Shellenberger and W. H. Ziemke. Dr. Shellenberger read the paper. Questioned by Messrs. Heald and Mitchell.

Paper—"Micro Baking—Technique, Applications and Results," by W. V. VanScoyk.

Paper—"Dough Mixing Studies," by J. Freilich and C. N. Frey. Read by Mr. Freilich. Discussion by M. D. Mize, V. C. Kylberg, and C. W. Brabender.

Paper—"Dough Development as Affected by Certain Mechanical and Chemical Treatments," by J. C. Baker and M. D. Mize. Mr. Mize read the paper. Discussion by Dr. Haase and Mr. Richards.

Paper—"The Relationship Between Protein Content and Flour Strength as Influenced by Additions of Malted Wheat Flour and Potassium Bromate," by T. R. Aitken, W. J. Eva, and W. F. Geddes, presented by Dr. Geddes.

Paper—"Further Investigations Into Nature of the Action of Bromates and Ascorbic Acid on the Baking Characteristics of Wheat Flour," by H. Jorgensen. This paper was read by W. F. Geddes. Discussion by Dr. Read.

Chairman Bohn recessed the meeting for lunch at 1 p.m.

The Wednesday afternoon session opened at 2:12 p.m. C. H. Bailey called the meeting to order and then turned the meeting over to the presiding chairman, Betty Sullivan. She immediately called on Dr. Cathcart.

Paper—"A Practical Method of Obtaining Permanent Records of Baking Studies—Characteristics of Crumb and Texture Are Effectively Recorded," by W. H. Cathcart.

Paper—"The Chemistry of Starch," by C. S. Hudson. This paper was presented in masterful style by our guest speaker and created a great deal of interested and interesting discussion.

Paper—"Laboratory Shortcuts and Gadgets," by J. T. Flohil.

Paper—"A Collaborative Study on Sugar Methods," by F. C. Hildebrand.

Paper—"A Collaborative Study of the 15-Minute Flour Moisture Method," by H. W. Putnam. Discussion by M. C. Markley, C. F. Davis, R. M. Sandstedt, and Perie Rumold.

Paper—"A Rapid Method for the Determination of Wheat and Flour Pigments," by D. S. Binnington.

Paper—"The Adaptation of the Pressuremeter to the Testing of Yeast," by R. T. Bohn. Discussion held over until after the next paper.

Paper—"The Pressuremeter Evaluation of Malt Products," by C. F. Davis and H. E. Tremain. Discussion by R. M. Sandstedt and R. T. Bohn.

Paper—"The Relation of Alpha and Beta Amylases to the Evaluation of Malts for Baking Purposes," by R. M. Sandstedt.

Paper—"Comparison of Various Types of Ash Dishes," by W. J. Eva, Nancy Milton, and W. F. Geddes. Dr. Geddes read the paper. Discussion by F. C. Hildebrand.

Paper—"General Report of the Committee on Methods of Analysis," by R. M. Sandstedt. This report was delayed for acceptance until the Friday session.

Dr. Sullivan recessed the meeting until Thursday morning. The recess came at 5:25 p.m.

Wednesday evening at 6:30 the annual banquet was held in the Pavillon Caprice at the Netherland Plaza Hotel. At this banquet Dr. Swanson received the Osborne Medal. Following this award, we were entertained by an excellent floor show and dancing.

Thursday, May 26

President Bailey called the meeting to order at 9:15 a.m. He then read a communication from C. C. Fifield telling us of the death of his father. Dr. Bailey appointed J. M. Doty and R. C. Sherwood to send a message of sympathy and flowers. He then called on Dr. Malloch.

Paper—"Further Studies with a Recording Mixer for Use with Small Samples," by J. G. Malloch. Discussion by C. H. Bailey.

Paper—"Relationship of Mixing Speed to Dough Development," by O. Stamberg and C. H. Bailey. Dr. Stamberg read the paper. Discussion by Washington Platt, C. H. Bailey, Oscar Skovholt, and C. W. Brabender.

Paper—"A New Method and Apparatus for Testing Doughs," by J. C. Baker. Discussion by G. Moen, W. Platt, C. B. Kress, O. Skovholt, and C. H. Bailey.

Paper—"The Theory of the Colloidal Structure of Dough as a Means of Interpreting Quality in Flour," by C. O. Swanson.

Paper—"Sugars in Bread," by Whitman Rice.

Paper—"A Summary of Biometrical Terms of Particular Application to Cereal Chemists." A committee report of the Committee on Definition of Technical Terms, by C. L. Brooke. Final action on this report was deferred until the Friday business session.

At 12:35 p.m. the meeting was recessed for lunch. A luncheon was held in the Pavillon Caprice for installation of new officers. The following motion was unanimously passed by the A. A. C. C. at this President's luncheon:

> Whereas the American Association of Cereal Chemists has just concluded another highly successful year in which it has progressed scientifically and in other ways, and
>
> Whereas during this year the Association has enjoyed the leadership of one of its most distinguished members as President, namely Dr. Clyde H. Bailey, and
>
> Whereas Dr. Bailey has in the past served the Association most generously in many capacities,
>
> Be it now resolved, that this Association hereby express its appreciation of the outstanding services of Dr. Bailey especially during his year as President;
>
> And be it further resolved that the incoming President be requested to appoint a committee to present to Dr. Bailey a permanent token as an indication of the affectionate esteem of the members.

The meeting was reopened at 2:35 p.m. Dr. Bailey introduced H. V. Moss, the presiding chairman for this session. Mr. Moss then called on J. W. Read for the first paper.

Paper—"Further Studies on the Activation and Inhibition of Flour Proteinase," by J. W. Read and L. W. Haas. Dr. Read read the paper.

Paper—"The Preparation and Properties of Wheat Proteinase," by A. K. Balls and W. S. Hale, read by the latter. Discussion by Dr. Read and M. D. Mize.

Paper—"Chlorophyll and Photosynthesis," by O. L. Inman. Dr. Inman was a guest speaker and his paper created a great deal of interest. Questioned by J. G. Malloch and Paul Logue. Chairman Moss thanked Dr. Inman.

Paper—"Action of Amylases on Raw Wheat Starches," by O. Stamberg and C. H. Bailey.

Paper—"A Histological Study of the Degradation of Wheat Starch by Amylase," by S. Redfern and W. Schanzenbach. Dr. Redfern read the paper. Discussion by M. J. Blish.

Paper—"A New Electro-Dialyzer for the Preparation of Beta Amylase," by S. Redfern.

Paper—"A Convenient Apparatus for Blish Manometric Gassing Power Determination," by J. G. Malloch. Questioned by R. M. Sandstedt.

Paper—"Chromotographic Fractionation of the Carotenoid Pigments of Flour," by C. H. Bailey and A. Houk. Dr. Bailey read the paper.

The Thursday afternoon session was closed at 5:25 p.m.

Friday, May 27

Dr. Bailey called the meeting to order at 9:20 a.m. and then turned the meeting over to the presiding chairman, R. A. Barackman.

(Preceding this session, the officers and local section chairmen were the guests of the Cincinnati Section at an 8 o'clock breakfast. Those present included M. C. Markley, P. R. Pitts, Martin Wise, C. F. Davis, G. F. Garnatz, R. A. Barackman, Perie Rumold, J. W. Montzheimer, W. F. Cathcart, Loren Francis, H. D. Liggitt,

Jr., F. R. Schwein, E. B. Brown, G. L. Alexander, Walter Reiman, Victor E. Marx, Whitman Rice, H. C. Gore, Clarence Oppen, Oscar Skovholt, John Shellenberger, C. H. Bailey, W. F. Geddes, and J. M. Doty. A very interesting discussion of matters pertaining chiefly to local section administration followed.)

Chairman Barackman announced that the papers by Dr. Barmore and Dr. Dunn would be moved up from the afternoon session to the morning session to save time and to present them along with other papers following the same line of thought. He then called on Pearl Brown.

Paper—"Report of the Sub-Committee on Methods of Testing Biscuit and Cracker Flours," by Miss Brown. Questioned by Dr. Brown.

Paper—"Report of Sub-Committee on Methods of Testing Self-Rising and Phosphated Flours," by O. E. Goolsms. Read by C. C. Walker.

Paper—"Report of a Convenient Crumb Color Standard for Self-Rising Flour," by G. W. Pearcy.

Paper—"A Study of Definitions and Various Items on the Score Card," by E. McKim and H. V. Moss, read by the latter. J. A. Dunn brought up the matter of the meaning of crumb and texture. M. C. Markley suggested that this should be a matter for the Committee on the Definition of Terms. H. W. Putnam suggested that the Soft Wheat Committee would be glad to work together with any other committees and try to clarify this point. He said he would welcome the cooperation of the Terms Committee or any individual suggestions.

Paper—"Report of Sub-Committee on Methods of Testing Cake Flours," by J. W. Montzheimer. Discussion by C. F. Davis, C. B. Kress, and L. D. Whiting.

Paper—"Adjustments Necessary to Compensate for Changes in Altitude When Baking Cakes," by M. Barmore. Read by L. D. Whiting. Discussion by R. W. Mitchell, W. R. Green, Mr. Smith, C. H. Bailey, W. E. Stokes, and J. W. Montzheimer.

H. W. Putnam moved the acceptance of the report on Methods of Testing Biscuit and Cracker Flours; seconded by Paul Logue; carried.

H. W. Putnam moved the acceptance of the Report on Methods of Testing Self-Rising Flours; seconded by R. C. Sherwood; carried.

H. W. Putnam moved the acceptance of the report on Methods of Testing Cake Flours; seconded by M. C. Markley; carried.

Paper—"The Leavening Action of Air Included in Cake Batter," by J. A. Dunn and J. R. White, read by the former.

Mr. Barackman introduced R. A. Kehoe.

Paper—"The Sources and Fate of Lead in Human Food," by R. A. Kehoe. Discussion by Paul Logue, C. H. Bailey, L. Zeleny, J. W. Read, and O. Skovholt.

Chairman Barackman then turned the meeting back to Dr. Bailey who called a recess until 2 p.m.

Dr. Bailey called the afternoon session to order at 2:10 p.m. and introduced the presiding chairman, H. M. Simmons, who introduced C. W. Brabender.

Paper—"New Experiences on the Dosage of Oxidizing and Bleaching Agents," by C. W. Brabender. Discussion by W. F. Geddes, J. W. Read, and J. G. Malloch.

Paper—"Maltose Fermentation Activators as Affecting Baking," by R. M. Sandstedt and M. J. Blish, read by Mr. Sandstedt. Discussion by M. C. Markley and C. H. Bailey.

Paper—"A Combined Photoelectric Colorimeter and Color Analyzer for Cereal Produts," by D. S. Binnington and P. A. McDonald. Read by D. S. Binnington. Questioned by R. C. Sherwood.

Paper—"Acidity in Cereals and Cereal Products," by L. Zeleny and D. A. Coleman, read by L. Zeleny. Questioned by M. C. Markley and R. C. Sherwood. It was suggested that copies of this paper be sent to the Secretary and then distributed to the members of the Association.

Chairman Simmons turned the meeting back to President Bailey for the business session. R. M. Sandstedt moved the acceptance of the Report of the Methods of Analysis Committee; seconded by M. D. Mize; carried.

C. H. Bailey moved the acceptance of the report of the Malt Analysis Standardization Committee; seconded by M. D. Mize; carried.

Quick Landis moved the acceptance of the report of the Committee on Definitions of Technical Terms; seconded by J. Freilich; carried.

Dr. Bailey then called Mr. Garnatz forward to thank him for the excellent meeting. He also called Walter Reiman forward to thank him. Mr. Reiman then

called on those present who had served on the Local Arrangements Committee to come forward. He introduced F. Schwein, J. A. Shellenberger, G. F. Garnatz, F. J. Coughlin, and Mrs. Oppen. Mrs. Oppen thanked the members for coming and asked them all to come again. Mr. Reiman also thanked the Association for coming to Cincinnati.

M. C. Markley thanked the men who had done such an excellent job of projecting the various slides and paper sheets. He suggested that all members who wanted to present papers in the future be made to realize the importance of having their slides made on transparent glass of standard size to assist these men in making good projections.

A motion was made to the effect that in the future all those appearing on this program be strongly urged to have all projections made up on standard transparent slides; seconded by O. Skovholt; carried.

C. H. MacIntosh read the report of the Resolutions Committee and moved its acceptance; seconded by R. C. Sherwood; carried. Report on page 568.

President Bailey then gave a short farewell speech and turned the gavel over to President-elect W. F. Geddes. Dr. Geddes read the names of the chairmen of the various committees.

M. D. Mize then moved that this Twenty-Fourth Annual Meeting be adjourned; C. H. MacIntosh seconded the motion; carried. Adjourned at 4:20 p.m.

Report of the Executive Committee

W. F. Geddes, *Chairman*

As defined in the Constitution, the Executive Committee is charged with the responsibility of carrying on the business activities of the Association and deciding upon matters of policy between meetings. The preparation of a budget has greatly facilitated the control and proper classification of Association expenditures and the year under review has been a prosperous one financially.

The Convention Reserve Fund has been brought to $1,000 through allocation of part of the unexpectedly large surplus arising from the Minneapolis meeting last year. The balance of this surplus was allotted to CEREAL CHEMISTRY for the purpose of facilitating more prompt publication of the papers presented at the annual meeting.

The assets of the Benefit Building and Loan Association of Kansas City, in which $2,000 of our funds were invested in 1926, have been assumed by the North American Savings and Loan Association, Kansas City, and our Association now holds Class A stock with a face value of $400 drawing interest and Class B certificates representing $1,600 of Class B assets in the latter company. At present it is impossible to report what returns may be expected from this investment.

Our Association was requested to place an exhibit in the exposition held in connection with the Kansas City national meeting of the American Bakers' Association last October. The Kansas City Section assumed the responsibility of making the exhibit. Many favorable comments on the excellency of the exhibit have been received and your Committee takes this opportunity of thanking the Kansas City Section for its valuable and capable services in this connection.

As has been the custom in the past, the Committee held a mid-year meeting in Chicago during the convention of the American Association of Bakery Engineers; at the invitation of this organization representatives of our Association participated in the program and Oscar Skovholt and Ray Bohn were the principal speakers on our behalf.

In March the Committee approved the plans of the Local Arrangements Committee in connection with the Cincinnati meeting.

The Committee unanimously accepted the report of the Osborne Medal Award Committee recommending C. O. Swanson, professor and head of the Department of Milling Industry, Kansas State Agricultural College, as the next recipient of the Thomas Burr Osborne Medal and authorized the necessary appropriations for its presentation at the Cincinnati meeting.

Invitations were received from the Kansas City Section and the Chamber of Commerce to hold the 1939 meeting in Kansas City. This invitation was especially urgent since the year 1939 will mark the twenty-fifth anniversary of the founding of the Association in that city. In view of this, and because the next convention is logically due to be held in the southwest, your Committee has selected Kansas City for the meeting in 1939.

In the death of D. A. Coleman on February 25, 1938, the Association lost a respected and valued member. He ably filled the post of Editor-in-Chief of CEREAL CHEMISTRY since 1931 and your Committee approved the appointment of M. J. Blish to complete his unexpired term of office. Dr. Coleman's death also left vacant the chairmanship of the Malt Analysis Standardization Committee and Elsie Singruen accepted this post.

The Executive Committee has given preliminary consideration to the feasibility of providing a business manager. The Association is approaching a size where the routine duties of certain officers have become too burdensome for them to undertake in their private capacities. This situation could be rectified and the business of the Association facilitated by the appointment of a business manager. Thus, the Association might combine in this office certain responsibilities of the Treasurer (in the matter of sending out invoices, collecting dues and keeping the books) with those of the Managing Editor of CEREAL CHEMISTRY in regard to soliciting advertising, keeping track of subscriptions and the numerous accounts incidental to the operation of the journal. The Association would appear to be financially able to pay a reasonable stipend, for at least part-time employment, to someone qualified to handle these purely business activities. As any action is contingent upon future arrangements for the publication of CEREAL CHEMISTRY, your Executive Committee recommends that the incoming Committee should give careful consideration to the proposal to appoint a Business Manager.

It appears that much useful work could be done and the position of cereal chemists greatly strengthened by closer co-operation with trade associations such as the Bakery Engineers and Operative Millers. Your executive, therefore, suggests that the advisability of appointing a special committee on co-operative research be taken under consideration by the incoming president to work with these and similar organizations on projects involving chemical problems.

At present, the local sections hold their annual meetings at various times of the year. It would be highly desirable for these to be held at the last section meeting preceding the national meeting so that the new officers would be installed prior to the convention, and it is hoped that the various local sections will find it convenient to conform to this suggestion.

The chairman wishes to express his appreciation to Dr. Bailey, who acted as chairman during his absence overseas, to the other members of the Committee, and the Secretary and Treasurer, for their help and co-operation.

Report of the Secretary

J. M. Doty

The work of the Secretary during the past year has been chiefly routine. It has consisted of writing letters of appreciation to workers and speakers at the last Annual Meeting, notifying all committee members of their appointments, caring for the general correspondence of the Association, contacting the officers of the Local Section, writing letters of welcome to new members and seeing that they get in touch with their nearest Local Sections, assembling and preparing some material for the News Letters, and maintaining an up-to-date membership list for the use of members.

Report of the History Committee

Rowland J. Clark, *Chairman*

The history of the Association has been written up to the present administration. This has been accomplished in two ways. First, the past presidents were requested briefly to outline the events of their respective terms in office. All but two responded very promptly. Second, the committee itself wrote up the administrations which were lacking.

As this history is studied it reveals not only the progress made by the Association, but also the changing tone of thoughts held by cereal chemists toward their profession. Ten years ago physical tests as applied to bread doughs were college curiosities. Today many laboratories include some form of recording dough mixer curves as a routine test. Ten years ago many chemists were using the recommended A. A. C. C. baking test formula and procedure. Today very few use this test. New ideas of today give way to discoveries of tomorrow.

Report of the Inter-Relations Committee

R. C. Sherwood, *Chairman*

The Committee continued its activities along the lines previously reported. Invitations to the annual meeting at Cincinnati and reprints of the program were sent to a large number of executives of milling, baking, malting and brewing companies. Many replies were received expressing continued interest in A. A. C. C. affairs.

It is recommended that the Inter-Relations Committee be continued with similar duties, with particular attention to efforts to cement good will between the A. A. C. C., allied associations and the food industries. It is suggested that the Committee again aid in distributing to interested persons the printed reports of the presentation of the Osborne Medal Award to C. O. Swanson. In addition, the Committee should take advantage of opportunities to distribute reprints, reports or notices of other important activities of the Association as a means of giving dignified publicity to accomplishments of the Association and its members.

Report of Committee on Membership Requirements

R. C. Sherwood, *Chairman*

This Committee was appointed a little more than a year ago to study the requirements for membership in the A. A. C. C. A report of progress was made at the 1937 meeting and subsequently published in the July issue of CEREAL CHEMISTRY. The Committee was reappointed to continue the study.

During the past year the problem has been discussed further by the Committee by correspondence. It was not possible to assemble the Committee until the Cincinnati meeting, at which time a proposal was drafted for consideration by members of the Association in the business session. The suggested revisions of the Constitution include:

(1) Raising the qualifications for membership somewhat, and defining them more clearly;
(2) Modifying the corporation membership to admit individuals as well as corporations;
(3) Establishing a membership application committee to relieve the Treasurer and the Executive Committee of the duty of reviewing applications.

Under item (1) the Committee suggests that the clause in the Constitution covering active membership be revised to read substantially as follows:

ARTICLE III

Section 2. The active membership of this association shall be restricted to—

(*a*) Those persons having the degree of Bachelor of Science (or equivalent) with a major in science from a college, university or technical school of recognized standing, or having at least four years of collegiate training with a major in science in such an educational institution.
(*b*) Those presenting evidence that they have had at least two years of training in chemistry, plus four years of practical experience in a chemical laboratory involving varied chemical and technological determinations.

Under item (2) the following revision is suggested:

ARTICLE III

Section 6. Corporations or individuals who are interested in or concerned with the use of cereals or cereal products may become sustaining members upon application to the Treasurer.

ARTICLE VI

Section 5 (in part). Sustaining members and their representatives shall have the privilege of attending all general meetings and in addition the privilege of the floor, but shall have no vote.

Under item (3) a new amendment substantially as follows is suggested:

ARTICLE IV

Section 8. The President shall appoint three active members, two of whom shall be past national officers, as a Membership Application Committee, the duties of which shall be the examination of applications for membership with full authority to approve or disapprove all applications submitted to it by the Treasurer. The Committee shall notify the Treasurer who shall inform the candidate of the disposition of his application.

ARTICLE III

Section 4. Applications for membership accompanied by the proper fee shall be directed to the Treasurer who in turn shall refer them to the Membership Application Committee, its decision to be final and upon report to the Treasurer to be transmitted by the Treasurer to the candidate.

The proposed revisions can be compared with the present Constitution by reference to CEREAL CHEMISTRY, volume 13, pages 492–495 (1936).

None of the above proposed revisions will affect those who are now members of the Association.

Five members of the Committee approved the proposed revision under item (1), the sixth member dissenting on the grounds that the present membership requirements are amply restrictive. All members of the Committee approved the proposal in item (2). Five members of the Committee favored the proposal in item (3), the sixth member not voting.

Serious consideration was given by the Committee to the establishment of associate membership but after extended debate, members of the Committee voted against this form of membership.

The above suggestions for revision of the Constitution are presented by the Committee with the request that the President ask for an expression of opinion concerning the issues involved for the guidance of the next Committee on Membership Requirements. The recommendations of the Committee are presented for informal discussion, not as the final form of amendments for adoption. Modification of membership requirements is an important issue that deserves careful consideration by all members of the Association. If the majority of the members of the Association are in favor of the recommendations substantially as presented, the Committee on Membership Requirements may then be instructed by the officers to proceed with the task of revising the many sections of the Constitution that must be changed to conform with the modifications in requirements.

Report of the Committee on Osborne Medal Award

Paul Logue, *Chairman*

The Committee has unanimously recommended Charles Oscar Swanson as the next recipient of the Thomas Burr Osborne Medal, awarded by the American Association of Cereal Chemists for "distinguished contributions in cereal chemistry."

Report of the Investment Committee

Paul Logue, *Chairman*

No funds have been reported by the Treasurer as available for investment during the current year and no specific investment recommendation has been made.

It is recommended that the amount of funds held in savings accounts be not less than 10% nor greater than 25% of the annual receipts.

It is recommended that investment of surplus funds be made only in legally authorized investments for savings banks and trust funds in the State of New York. A copy of the laws limiting the investments in savings banks in New York State has been made a part of the original of this report.

Report of the Publicity Committee

Victor E. Marx, *Chairman*

The Publicity Committee has prepared a list of recognized business publications in the milling, baking, brewing, malting, food, and general science fields, and supplied each section with a copy of this list for publicity purposes.

The Publicity Committee has cooperated with the local publicity chairmen by directing their attention to the authorized list of publications.

We have prepared for display a partial exhibit of clippings obtained for the cereal chemists' activities in some of the papers served by this Committee.

One of the things which the Publicity Committee hopes to do through its activities is to enhance the prestige and standing of chemists in their own communities, by "selling" chemists to industry.

We hope to perfect a better organization during the coming year, in order to operate more efficiently.

Closer contact is needed between the Publicity Committee and other committees of the Association in order that we may develop material covering their activities suitable for publicity purposes.

Report of the Traffic Committee

G. Norman Bruce, *Chairman*

The Traffic Committee has little to report. The Chairman wishes to express his indebtedness to his active and efficient committee members, Messrs. Clark, Clulow, Gregory, Markley, Tibbling, and Vernon, for really doing the work.

The Chairman suggests that the Traffic Committee be made a part of the Convention Arrangements Committee and that the chairman be appointed from the convention city for convenience. Traffic arrangement is a natural part of a convention build-up and has a direct bearing on program arrangements.

Report of the Membership Committee

D. A. MacTavish, *Chairman*

The efforts of your committee have been directed not toward a classification of membership, educational qualifications, *etc.*, subjects which have been well canvassed in the past, but toward maintaining and, wherever possible, increasing the "active" membership of the local sections. This was done in the belief that without a growth in this class of membership, the interest in the national Association's welfare will tend to lag and become too localized.

We are pleased to report a total of 570 members in good standing, which represents *an increase in the active membership at the middle of May of 30 in excess of this time last year.* Actually, there has been an addition of 38 active and 4 corporation members—the difference between these figures and the net increase, as above, being made up by delinquents and resignations. We feel that this is a very creditable showing, considering the depressed and difficult economic conditions which have prevailed during the past year.

An incomplete survey of the first of May of the standing of "active" and "associate" membership, by sections, reveals:

In Active Membership

5 sections with an increase
2 sections with no change
4 sections giving no comparison with last year
1 section with a decrease
1 section not heard from

In Associate Membership

3 sections report an increase
3 sections report no change
4 sections give no comparison
1 section reports a decrease
1 section not heard from

Due to variety of interests and type of programs presented, some sections cater to a much larger associate membership than others and it is essential that they should so continue if sustained interest in local section activities is to be maintained. This puts them out of balance as far as the 75–25 proportion of "Active" to "Associate" membership (provided in our constitution) is concerned. Nevertheless, we feel that provided these sections maintain a healthy and growing active membership, they are not to be censured because of a condition which is essential for their continued well being.

In conclusion, allow me to thank the members of the committee and in particular the chairmen of the local sections for their kindly and effective co-operation during the past year without which the above showing would not have been attained.

Report of the Committee on Resolutions

C. H. MacIntosh, *Chairman*

Whereas, the American Association of Cereal Chemists has again experienced a most successful annual meeting, and

Whereas, this success has in a large measure been due to the splendid co-operative services rendered by the officers and committees of the Association.

Therefore be it resolved that the most sincere appreciation of this Association be expressed to the officers who have served during the past year: President, C. H. Bailey; Vice-President, W. F. Geddes; Secretary, J. M. Doty; Treasurer, Oscar Skovholt; and to the Program Committee, G. F. Garnatz, Chairman; the Local Arrangements Committee, Walter Reiman, Chairman; the Ladies' Entertainment Committee, Mrs. C. O. Oppen, Chairman, and to all other Committees who, by their contributions, have made the year's work so successful.

Be it further resolved that we express our appreciation for the services of the late D. A. Coleman, Editor-in-Chief of CEREAL CHEMISTRY; and that we express our appreciation to M. J. Blish who has served as Editor-in-Chief since the untimely passing of Dr. Coleman; be it further resolved that we sincerely thank C. C. Fifield, Managing Editor of CEREAL CHEMISTRY, and his associates on the business and editorial staff of CEREAL CHEMISTRY for their splendid work during the year just passed.

Be it further resolved that we express thanks to Rev. L. W. Almy, who delivered the invocation at the opening session.

Be it resolved that we express our thanks to the Hon. James G. Stewart, Mayor of Cincinnati, for the fine welcome extended the Association on behalf of the City of Cincinnati, and to the Hon. Russell Wilson, member of the Council of the City of Cincinnati, for his fine address at the luncheon to install the new officers.

Whereas, the success of these meetings has been due in no small measure to the inspiring and instructive contributions of the guest speakers who appeared on our program,

Therefore be it resolved, that we, through the offices of our Secretary, express our most sincere thanks and appreciation to H. G. Knight, Chief of the Bureau of Chemistry and Soils, United States Department of Agriculture; and L. E. Caster, representing the American Society of Bakery Engineers; B. B. Bayles, United States Department of Agriculture; W. W. Worzella, Purdue University; Ray Sopher, Superintendent, Acme Evans Milling Co.; G. S. Sperti, Director of Research, Institutum Divi Thomae; J. R. Loofbourow, Institutum Divi Thomae; E. S. Cook, Institutum Divi Thomae; C. S. Hudson, National Institute of Health; W. B. Newkirk, Corn Products Refining Co.; A. K. Balls and W. S. Hale, Bureau of Chemistry and Soils, United States Department of Agriculture; O. L. Inman, Director of the C. F. Kettering Foundation, Antioch College; R. A. Kehoe, Director of the Laboratory of Applied Physiology, University of Cincinnati; H. Jorgenson, Copenhagen, Denmark.

Be it further resolved that we express our sincere thanks and appreciation to the following, whose assistance and co-operation with the Local Arrangements Committee have contributed materially to the success of the many and varied features of our meeting: Lawrenceburg Roller Mills; American Agricultural Chemical Co.; Monsanto Chemical Co.; Laboratory Construction Co.; Lever Bros. Co.;

Precision Scientific Co.; Durkee's Famous Foods; Victor Chemical Works; Wallace and Tiernan Co.; Kroger Food Foundation; Kroger Grocery and Baking Co.; Proctor and Gamble Co.; American Soya Products Co.; The Cincinnati Post; The Cincinnati Times-Star; Cincinnati Enquirer; the Burger Brewing Co.; Hudepohl Brewing Co.; Cincinnati Scientific Co.; John Shillito Co.; Mr. Royal Ryan, Sales Manager of the National Hotel Management Co.; the Netherland Plaza Hotel; Jack Rockaway, Cincinnati Chamber of Commerce.

Whereas during the year, six of our members have passed on to their reward, Therefore be it resolved, that we express our deep regret at the passing of D. A. Coleman, E. E. Werner, J. R. Katz, Charles J. Henry, Mr. Hahn, and O. B. Winter. And be it further resolved that the Secretary be instructed to convey to their families the deep and heart-felt sympathies of the Association.

A. A. C. C. Committees

Executive Committee

G. F. Garnatz, *Chairman* — C. H. Bailey — H. D. Liggitt, Jr.
W. F. Geddes — C. F. Davis

Membership Committee

D. A. MacTavish, *Chairman* — Bert Ingels — J. M. Doty
Chairmen of local sections

Committee on Methods of Analysis

R. M. Sandstedt, *Chairman* — F. C. Hildebrand — W. L. Heald
H. W. Putnam — D. S. Binnington — R. F. Barackman

Malt Analysis Standardization Committee

Elsie Singruen, *Chairman* — H. W. Rohde — H. C. Gore
G. S. Bratton — A. D. Dickson — J. A. Anderson
C. Rask

Committee on Definitions of Technical Terms

Quick Landis, *Chairman* — Washington Platt — Clinton Brooke

Committee on Employment

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